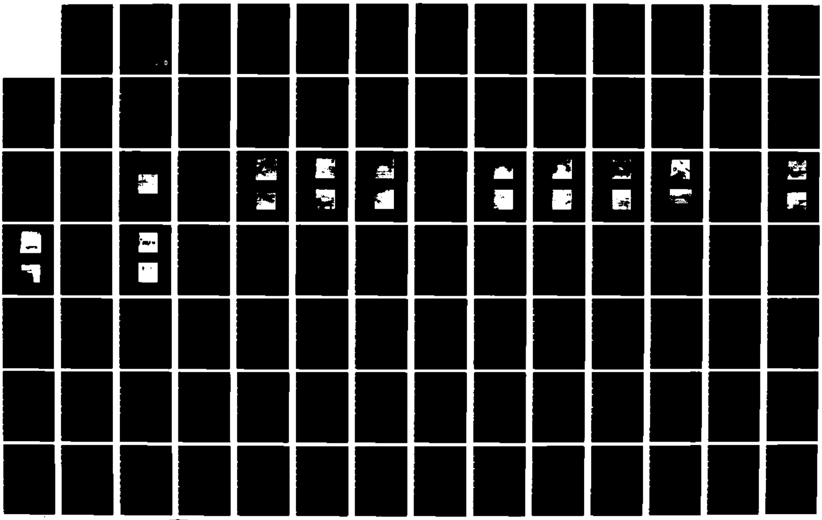
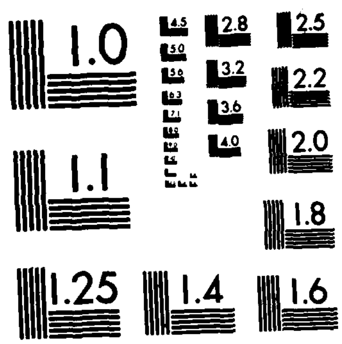
AD-A128 506 MOSQUITO PRODUCTION AND ASSOCIATED ENVIRONMENTAL AND CHEMICAL PARAMETERS OF BREEDING WATERS(U) AIR FORCE INST OF TECH WRIGHT-PATTERSON AFB OH T J KASA MAY 83 1/2

UNCLASSIFIED AFIT/CI/NR-83-13T F/G 6/3 NL





MICROCOPY RESOLUTION TEST CHART
NATIONAL BUREAU OF STANDARDS-1963-A

UNCLASS

SECURITY CLASSIFICATION OF THIS PAGE (When Data Entered)

REPORT DOCUMENTATION PAGE

READ INSTRUCTIONS BEFORE COMPLETING FORM

1. REPORT NUMBER AFIT/CI/NR 83-13T	2. GOVT ACCESSION NO. A128506
3. RECIPIENT'S CATALOG NUMBER	
4. TITLE (and Subtitle) Mosquito Production and Associated Environmental and Chemical Parameters of Breeding Waters	5. TYPE OF REPORT & PERIOD COVERED THESIS/~~DISSERTATION~~
	6. PERFORMING ORG. REPORT NUMBER
7. AUTHOR(s) Thomas Joseph Kasa	8. CONTRACT OR GRANT NUMBER(s)
9. PERFORMING ORGANIZATION NAME AND ADDRESS AFIT STUDENT AT: Purdue University	10. PROGRAM ELEMENT, PROJECT, TASK AREA & WORK UNIT NUMBERS
11. CONTROLLING OFFICE NAME AND ADDRESS AFIT/NR WPAFB OH 45433	12. REPORT DATE May 1983
	13. NUMBER OF PAGES 116
14. MONITORING AGENCY NAME & ADDRESS(if different from Controlling Office)	15. SECURITY CLASS. (of this report) UNCLASS
	15a. DECLASSIFICATION/DOWNGRADING SCHEDULE

16. DISTRIBUTION STATEMENT (of this Report)

APPROVED FOR PUBLIC RELEASE; DISTRIBUTION UNLIMITED

17. DISTRIBUTION STATEMENT (of the abstract entered in Block 20, if different from Report)

18. SUPPLEMENTARY NOTES

APPROVED FOR PUBLIC RELEASE: IAW AFR 190-17

Lynn Wolaver 17 May 83

LYNN E. WOLAVER
Dean for Research and
Professional Development
AFIT, Wright-Patterson AFB OH

19. KEY WORDS (Continue on reverse side if necessary and identify by block number)

20. ABSTRACT (Continue on reverse side if necessary and identify by block number)

ATTACHED

DTIC
SELECTE
MAY 2 6 1983
S D
E

AD A128506

DTIC FILE COPY

DD FORM 1 JAN 73 1473 EDITION OF 1 NOV 65 IS OBSOLETE

UNCLASS

SECURITY CLASSIFICATION OF THIS PAGE (When Data Entered)

ABSTRACT

Kasa, Thomas Joseph. M.S., Purdue University, May 1983. Mosquito Production and Associated Environmental and Chemical Parameters of Breeding Waters. Major Professor: Gary W. Bennett.

Larval surveillance techniques were employed to identify mosquito breeding sites in the vicinity of Lafayette, Indiana, and Purdue University. Emphasis was placed on the analyses of breeding waters containing *Aedes triseriatus* (Say), *Ae. vexans* (Meigen), *Culex pipiens pipiens* Linnaeus, *Cx. salinarius* Coquillett, and *Cx. restuans* Theobald because of their actual or potential disease vector importance. Other mosquito species collected during this study included *Anopheles punctipennis* (Say), *Cx. territans* Walker, *Culiseta inornata* (Williston), *Orthopodomyia signifera* (Coquillett), *Psorophora columbiae* (Lynch Arribalzaga), and *Ps. howardii* Coquillett.

A number of physically and environmentally different breeding sites were chosen for study. The 3 general breeding category types included artificial containers, ground cavities, and storm sewer catch basins. A total of 20 study sites were monitored. These breeding sites were analyzed as to species presence and relative abundance, general environmental setting, and selected water chemistry variables.

Cx. restuans was the most abundant species collected followed by *Cx. salinarius*, *Ae. triseriatus*, and *Cx. pipiens*. *Ae. triseriatus* was collected only from the artificial container habitat, while the 3 *Culex* species were collected from all monitored habitat types.

Multiple linear regression analyses determined pH and water temperature to be the most significant variables pertaining to overall mosquito development. The storm sewer catch basin habitat type was concluded to be the most important, with water temperature and magnesium the most significant of the analyzed variables. Physical construction of storm sewer catch basins also influenced mosquito production.

Numerous variable interactions resulting from multiple linear regression analyses are discussed for each site type, as well as for the major individual species. Ranges and means for the selected water chemistry variables are also reported and reflect the chemical parameters associated with the major study species. This information offers a comparative method for the reasonable prediction of species breeding in similarly analyzed habitats.

Accession For
NTIS GRA&I X
DTIC TAB
Unannounced
Justification
By
Distribution/
Availability Codes
Dist | Avail and/or Special
A
DTIC COPY INSPECTED 2

3-3

MOSQUITO PRODUCTION AND ASSOCIATED ENVIRONMENTAL AND CHEMICAL PARAMETERS OF BREEDING WATERS

A Thesis

Submitted to the Faculty

of

Purdue University

by

Thomas Joseph Kasa

In Partial Fulfillment of the

Requirements for the Degree

of

Master of Science

May 1983

83 0[illegible] 26 . 026

ACKNOWLEDGMENTS

The author wishes to express his sincere appreciation to Dr. Gary W. Bennett, Department of Entomology, Purdue University for his adept guidance, encouragement, and monumental patience during the course of this endeavor. To Dr. Ralph E. Williams, Department of Entomology, Purdue University, and Dr. Michael J. Sinsko, Indiana State Board of Health, equally sincere appreciation is expressed for their timely council and helpful advise. Gratitude is also expressed to Dr. Ronald A. Ward, Medical Entomology Project, Smithsonian Institution for his valuable assistance in the confirmation of specimen identifications. Thanks to the many individuals of the United States Air Force Occupational and Environmental Health Laboratory for their timely laboratory analyses. Appreciation is also expressed to Dr. Virgil Anderson, Department of Statistics, Purdue University, for his initial guidance on the development of viable regression programs. Additionally, thanks to Ms. Sandy Lindsey for her professional management and personal attention to the many administrative aspects of this program.

Most sincere appreciation is expressed to my family, without whose sacrifice and support this project would not have been completed. Loving thanks to Gloria, Tom, and Kimberly for their devotion and continuing words of encouragement through the most difficult of times.

TABLE OF CONTENTS

LIST OF TABLES

Appendix Table Page

LIST OF FIGURES

INTRODUCTION

The mosquito has long been recognized as the single most important vector of disease in the world. Mosquitoes have been acknowledged as a problem in cities and towns for years, but too little attention has been paid to where they breed and which factors, if any, influence their breeding. Sites chosen for study included artificial containers, ground cavities, and storm sewer catch basins. The majority of these sites were selected from residential areas because of the close proximity to human habitats. Complete analyses of mosquito breeding sites in such populated areas is needed to gain necessary insight into this major problem.

This research, conducted in the Lafayette/West Lafayette area of Indiana, concentrated on the breeding sites of locally proven or suspected vectors of disease. These species included *Aedes triseriatus*, *Culex pipiens*, *Cx. salinarius*, and *Cx. restuans*. Even though emphasis was placed on these species, the breeding parameters of all species found during this study were analyzed for comparative purposes. Consideration was also given to the seasonal sequence of species found at a particular site and their association with one another.

The objectives of this study were threefold:

1. To identify, by means of larval surveillance techniques, established mosquito breeding sites.
2. To analyze selected environmental and water chemistry variables from

each study site in relation to the mosquito species collected and population levels present.

3. To determine the influence of selected water chemistry and environmental variables on the breeding of study species.

The variables selected were recognized to have the most influence on, or comprise the greatest portion of, natural waters. The data gathered, together with knowledge of the general environmental setting, provides the information needed to reasonably predict potential mosquito breeding and population levels in similarly analyzed habitats.

LITERATURE REVIEW

Studies on mosquito bionomics have historically centered on those species which are responsible for the transmission of human and animal diseases. Several proven vector species were identified from the Lafayette, Indiana area during this study (Table 1). With the effective reduction in the occurrence of dengue, malaria, and yellow fever in this country, the various encephalitis viruses rank as the most significant mosquito-borne diseases still in evidence.

Mosquito-borne Disease and Vector History

The most important mosquito-borne disease to occur in the Midwest region of the United States has been St. Louis encephalitis (SLE). Outbreaks of SLE have occurred in areas of the Midwest, to include Indiana, with increasing frequency since the 1933-1934 epidemic in St. Louis, Missouri (Lumsden 1959). Vast numbers of *Culex* mosquitoes were credited with probable involvement. Subsequent evidence of SLE activity in the Ohio-Mississippi River Basin occurred at Calvert City, Kentucky, in 1955 (Razenhofer et al. 1957, Hayes 1973). During the next 9 years, only sporadic cases of SLE were reported by the Center for Disease Control (CDC). In Indiana, minor outbreaks of SLE occurred in Gibson County in 1957, and in Vandenburgh and Warrick Counties in 1965 (Newhouse and Siverly 1966a).

Table 1. Biological data on medically important mosquitoes in the Greater Lafayette, Indiana area.

Mosquito Species	Larval Habitats[a]	Biting Activity[b]	Flight Range[c]	Diseases Vectored[d]
Aedes				
triseriatus	TH, AC	D	0.5-1 mi	CE
vexans	FW, GP, IP, SSCB	C, N	1-5 mi	CE, (EEE)
Anopheles				
punctipennis	AC, DD, GRP	C	1-2 mi	(M)
Culex				
pipiens pipiens	AC, SSCB, GRP	C, N	0.5 mi	SLE, (WEE), (VEE)
restuans	WP, GRP, DD, SSCB	C, N	1-2 mi	(EEE), (WEE)
salinarius	GP, LM, FS, SSCB	C, N	1-5 mi	(EEE)
Culiseta				
inornata	GRP, DD	C, N	1-2 mi	(WEE), (CE)
Psorophora				
columbiae[e]	IP, RF, GRP	C, N	1-5 mi	VEE

[a] AC = artificial container; DD = drainage ditch; FS = freshwater swamp; FW = flood water; GP = grassland pool; GRP = ground pool; IP = irrigated pasture; LM = lake margin; RF = rice field; SSCB = storm sewer catch basin; TH = tree hole; WP = woodland pool

[b] C = crepuscular; D = day; N = night

[c] Values given are estimates of normal flight ranges

[d] ( ) indicate secondary vectors, otherwise, primary vectors. CE = California encephalitis; EEE = eastern equine encephalitis; M = malaria; SLE = St. Louis encephalitis; VEE = Venezuelan equine encephalitis; WEE = western equine encephalitis

[e] Formerly Psorophora confinnis

Over 1,000 cases of SLE were reported in the United States in 1964 (Mack et al. 1967). Epidemics occurred in several states including Texas, Tennessee, Kentucky, Illinois, New Jersey, and Indiana. This widespread occurrence of SLE prompted disease investigations in Danville, Kentucky (Mack et al. 1967), Memphis, Tennessee (Sudia et al. 1967), McCleansboro, Illinois (Kokernot et al. 1967), and southwestern Indiana (Newhouse and Siverly 1966b). Mack et al. (1967) reported 3 isolations of SLE virus from Cx. pipiens taken from Danville, Kentucky. Four confirmed human cases of SLE were identified in Memphis, Tennessee, and 4 isolates of SLE virus were obtained from pools of mosquitoes of the Cx. pipiens quinquefasciatus complex (Sudia et al. 1967). Similar findings were reported during and investigation in McCleansboro, Illinois (Kokernot et al. 1967). Forty pools of Cx. pipiens complex mosquitoes, collected from the McCleansboro area, yielded SLE virus. Twenty-two human cases of SLE, including 2 deaths, were reported from the area.

The 1964 SLE outbreak in Indiana occurred in the Evansville-Booneville area of the state. Nineteen human cases of SLE were reported, of which 2 were fatal. The peak case incidence occurred during the first part of September (Marshall 1965). Mosquitoes of the Cx. p. quinquefasciatus complex were collected and examination of male terminalia showed about 98% of the specimens to be typical Cx. pipiens. A colony of Cx. pipiens was later established in the laboratory and was shown experimentally to transmit 1 of the Evansville SLE virus strains with considerable efficiency (Chamberlain et al. 1966). Since the recognized northern limit of Cx. p. quinquefasciatus is approximately at the latitude of southern Indiana, it can be concluded that Cx. pipiens is the major vector of SLE in central Indiana. This conclusion is further supported by the studies of Hayes (1973).

One human case of SLE was reported from Allen County, Indiana in 1970 (Siverly 1972). Sporadic cases of SLE occurred in 1971, 1973, and 1974. A major outbreak occurred in 1975, with 318 cases of SLE, and 8 cases of California encephalitis (CE) reported from Indiana. Tippecanoe County accounted for 8 of the SLE cases during that year. A lower disease incidence was evidenced from 1976 to the present. Overall, approximately 380 cases of SLE and 28 cases of CE have been reported from Indiana since 1970 (Lang et al. 1981). Locally, 1 unconfirmed human case of SLE was reported in a Lafayette, Indiana, man who frequently played golf at the Lafayette Country Club during the late summer of 1976. The reported September time-frame fits the peak case incidence period observed by Marshall (1965) during the 1964 SLE outbreak in southwestern Indiana.

The CDC reported 1400 cases of primary encephalitis from the United States in 1981. This figure is in excess of the 1976-1980 median of 1185 cases. Of the 1400 cases, 35% were from the east-northcentral region of the country. Ohio had the greatest incidence with 245 cases, followed by Indiana with 145, Michigan with 68, Wisconsin with 25, and Illinois with 9. Over 90% of the cases from Indiana were identified as SLE, with the remaining cases identified as CE.

Biology and Larval Habitats of Study Species

Indiana is divided into 8 major natural divisions (Lindsey et al. 1969) which represent graded differences among vegetational, floristic, edaphic, and physiographic features. Many of these features influence the various mosquito species which are found in these natural divisions. The vast majority of Tippecanoe County, to include the Lafayette area, lies in the Tipton Till Plain Beech Maple Division (Division V). The

division contains poorly drained but highly productive timber soils. Many small woodlots and timber tracts are also scattered throughout division V. A variety of mosquito fauna has been reported from this area (Siverly 1972).

Early preliminary studies on the mosquito fauna of Tippecanoe County were accomplished by Hart (1942) and Brooks (1947). These initial surveillance studies identified mosquito species common to Tippecanoe County. Additional County records were continuously reported by Siverly after that period and summarized in 1972 (Siverly 1972). This summary does not include Cx. salinarius from Tippecanoe County. However, Brooks (1947) reported 49 specimens of this species taken in light traps during 1942 and 1946.

The majority of natural Ae. triseriatus breeding in Indiana occurs in tree holes and stump holes in wooded environments (Siverly 1972). Additional breeding site selection is associated with these surrounding wooded environments. This fact is supported by the findings of Sinsko and Craig (1979) in their study on the dynamics of Ae. triseriatus populations. They determined, through recapture methods, that the adults of this species do no move across open areas from woods to woods. Ae. triseriatus is a primary vector of the LaCrosse (LAC) virus of the CE group in the north central United States (Loor and DeFoliart 1970). Investigations have also shown that LAC virus can be transmitted transovarially by the species (Watts et al. 1973). Biting activity is greatest in the evening, although females will attack during the day in wooded areas.

Larvae of Ae. vexans have been reported breeding in roadside ditches, woodland pools, and other similar ground cavities. This species is cosmopolitan in Indiana and can be considered the most important pest species in the state (Siverly 1972). Ae. vexans has been implicated in the transmission of CE and eastern equine encephalitis (EEE). Females will feed at any time of the day, but peak activity is at dusk and dawn (Knight and Henderson 1967).

The larvae of Anopheles punctipennis have been reported from streams, ponds, and various ground cavities. In past years, the species has been reported as an important malaria vector in California (Siverly 1972).

Larvae of Cx. pipiens can be found in a variety of artificial container breeding situations. Storm sewer catch basins are also common breeding sites (Carpenter and LaCasse 1955, King et al. 1960, Maddock et al. 1963). Cx. pipiens is considered the most significant vector in Indiana due to its historic involvement in the transmission of SLE.

Although Cx. restuans is not considered an important vector species in Indiana, it has been implicated in the transmission of EEE and western equine encephalitis (WEE) in other portions of the country. Larvae have been collected from a wide variety of artificial container and ground cavity breeding situations. It is most frequently found in association with Cx. pipiens and Cx. salinarius.

Cx. salinarius is a species which is probably cosmopolitan over the state of Indiana. It has been previously reported from ground cavities, freshwater swamps, and storm sewer catch basins. Although not considered an important vector species in Indiana at present, Cx. salinarius has been identified as a vector of EEE in other areas of the United States. It is frequently found in close association with Cx. pipiens and Cx. restuans.

Cx. territans breeds in a variety of ground cavity situations, as well as tree holes and streams. It feeds on cold-blooded animals and only rarely attacks man (Means 1965, McIver 1969). This species is of no medical importance in Indiana at the present time.

Culiseta inornata, a secondary vector of WEE and CE, is not considered an important vector in Indiana. Larvae can be found in a variety of situations including bogs, marshes, pools, and roadside ditches. Ground cavities are by far the most common source of this species.

Larvae of Orthopodomyia signifera breed primarily in artificial containers found in or near wooded areas. The species is often found in association with Ae. triseriatus. Or. signifera is of no medical importance in Indiana.

The remaining incidental species, Psorophora columbiae and Ps. howardii, are common to ground pool and roadside ditch habitats. While neither species is considered to be of medical importance in Indiana, Ps. columbiae is recognized as a major vector of Venezuelan equine encephalitis (VEE) in the southern United States. The females of both species are painful biters.

Larval Population Sampling

The primary concern in choosing a larval sampling method is to insure continuity in sampling from site to site over time. The detection and measurement of mosquito populations has proved easier by light trapping of adults than by larval sampling (Service 1976). Loomis (1959) concluded that even the best sampling methods lacked standardization and reliability on a large scale. His concern centered on the lack of meaningful data. A larval density study by Barton (1977) indicated that,

overall, the heaviest larval densities occurred in from 2.5-15 cm of water. Chubachi (1976) determined that the standard pint dipper was as equally effective for sampling artificial container habitats as it was for sampling larval densities in rice fields. A larval concentrator constructed by Earle (1956) is not used as often. The use of standardized sampling methods allows for continuity in larval surveillance.

Environmental and Chemical Analyses of Variables

Individual water chemistry variables were selected because of their known importance to waters of the types which were monitored. Information relating these variables to mosquito breeding has been reported in the literature to one degree or another.

Water temperature has been referred to as a factor which influences natural mosquito breeding. Kliewer et al. (1964) reported the effects of a laboratory temperature gradient experiment on the distribution of *Aedes* larvae. The experiment showed a difference in the preferred mean temperature between 2 *Aedes* species. The preferred mean temperature varied 3°C between the species. The average location within the gradient for *Ae. nigromaculis* was at 25.7°C, while *Ae. melanimon* was located at 22.5°C. According to Shriver and Bickley (1964), the optimum temperature range for embryonic development of *Cx. p. quinquefasciatus* is from 23.9-29.4°C, a range within which about 70% of the eggs hatch in about 30 hr.

pH is an accepted measure of the hydrogen ion activity in moles/l. Various pH ranges and means have been reported from mosquito breeding waters. Bast (1963) reported mean pH values of 4.81 and 4.11 from 2 mosquito breeding pools. A mean pH of 4.73 was also reported from a non-breeding pool. Hagstrum and Gunstream (1971) reported mean pH values

ranging from 7.2-7.8 for breeding sites which contained 8 species of mosquito larvae. pH ranges for individual species were reported by Petersen and Chapman (1969) in their study on tree hole, stump hole, and ground cavity breeding sites. The pH ranges varied according to the type of site monitored. Petersen and Chapman (1969) also reported the pH ranges of each type of breeding habitat they sampled. The pH range for tree holes was 5.5-9.9, the range for stump holes 6.0-8.9, and the range for ground cavities 5.7-8.3. pH values ranging from 7.7-9.1 for *Ae. vexans* breeding sites were also reported by Petersen and Rees (1966).

The only available dissolved oxygen (DO) relationships regarding mosquito breeding waters were reported by Bast (1963). The 2 breeding pools which he surveyed had mean DO values of 7.20 mg/l and 5.75 mg/l, respectively. The single non-breeding pool surveyed had a mean DO value of 9.84 mg/l. No observations were made to indicate the importance of these values in relation to mosquito breeding or non-breeding.

Chemical oxygen demand (COD) analysis indicates the quantity of oxidizable materials present in a water sample. In some instances, correlations can be made between COD and the biochemical oxygen demand (BOD). In natural waters, the COD value can be interpreted to be approximately 50% of the BOD value. This relationship becomes much less accurate for polluted waters. Smith (1969) reported BOD values in his study on mosquito breeding in oxidation lagoons. These BOD values were used only to provide descriptions of the study lagoons and were not related to individual species.

Analysis for specific conductance (conductivity) yields a measure of a water's capacity to convey an electric current. This value is related

to the total concentration of the ionized substances in the water when standardized and measured at 25°C. Mean specific conductance values, in micromhos/cm, were reported by Bast (1963) in his work on pool breeding mosquito species. He reported mean values of .375 and .435 micromhos/cm x 10^3 for the 2 breeding pools and a mean value of .581 micromhos/cm x 10^3 for the non-breeding pool. Bast suggested that the importance of specific conductance may lie in the amounts of component elements which contribute to these values. More definitive specific conductance data was reported by Petersen and Chapman (1969). They reported both ranges and averages for breeding site types and individual species. Specific conductance values in micromhos/cm x 10^3 from the 3 different site types included a range of .12-10.00 and an average of 2.00 for tree holes; a range of .10-1.60 and an average of .53 for stump holes; and a range of .10-1.20 and an average of .39 for ground cavities. The ranges and averages for individual species reported by Petersen and Chapman (1969) included *Ae. triseriatus* with a range of .11-4.10 and an average of .85; *Cx. restuans* with a range of .10-1.25 and an average of .54; *Cx. salinarius* with a range of .35-1.07 and an average of .65; *Cx. territans* with a range of .10-.11 and an average of .10; and *Or. signifera* with a range of .12-10.00 and an average of 2.02 micromhos/cm x 10^3. Petersen and Rees (1966) reported specific conductance values for *Ae. vexans* ranging from .50-4.80 micromhos/cm x 10^3 with an average of 2.26. Petersen and Willis (1971) demonstrated the effects of salinity on site selection by ovipositing tree hole mosquitoes. The mean conductivity of water in artificial tree holes was used as the vehicle for measurement of salinity. In the sites where larval development routinely occurred, the mean

conductivity for <u>Or. signifera</u> was 4.60 micromhos/cm x 10^3, while that for <u>Ae. triseriatus</u> was 3.20 micromhos/cm x 10^3. These results indicate <u>Ae. triseriatus</u> is generally restricted to habitats with lower salinity, while <u>Or. signifera</u> appears to have a broader tolerance.

The presence of calcium in water results from association with limestone, dolomite, gypsum, or gypsiferous shale. Calcium ranks fifth among the elements in order of abundance and a portion of the hardness in water is attributed to the presence of calcium. The other major element which contributes to water hardness is magnesium, the eighth most abundant element. Magnesium is a common constituent of natural waters. Concentrations may range from 0 to several hundred mg/l, depending on the source.

The hardness of water is a measure of the capacity of the water for precipitating soap. Soap is precipitated chiefly by the calcium and magnesium ions commonly present in water. It may also be precipitated by ions of other polyvalent metals such as aluminum, iron, manganese, strontium, and zinc. Because only calcium and magnesium are usually present in significant concentrations in natural waters, hardness is defined as a characteristic of water that represents the total concentration of these ions expressed as calcium carbonate ($CaCO_3$). When the hardness is numerically greater than the sum of the carbonate alkalinity, the amount of hardness that is equivalent to the total alkalinity is called carbonate hardness. The amount of hardness in excess of this is called noncarbonate hardness. When hardness is numerically equal to or less than the total alkalinity, all hardness is carbonate hardness.

Chloride is one of the major inorganic anions in water and wastewater. The chloride concentration is higher in wastewater than in raw

water because sodium chloride is a common article of diet and passes unchanged through the digestive system. High chloride concentrations in mosquito breeding waters may be an indicator of wastewater source pollution. Kardatzke (1981, 1982) demonstrated that the addition of varying amounts of calcium chloride ($CaCl_2$) to the larval medium of Aedes snowmelt mosquitoes was detrimental to immature survival. The addition of $CaCl_2$ ranged from 500-5000 mg/l. The majority of Aedes species tested showed a 0% survival rate at levels above 2500 ppm.

Sulfate, widely distributed in nature, may be present in natural waters in concentrations ranging from a few to several thousand mg/l. Similarly, sodium, the sixth ranking element in order of abundance, is present in most natural waters. Concentrations may range from slight, to the relatively high levels found in brines and hard waters softened by the sodium exchange process. The ratio of sodium to total cations is important, in that, soil permeability is harmed by a high sodium ratio. Soils with these high ratios display increased water retention characteristics, and soils with low ratios display poor water retention characteristics. It follows that mosquito breeding sites containing high levels of sodium in the substrate retain water for longer periods of time.

Although potassium ranks seventh among the elements in order of abundance, its concentration in natural waters seldom exceeds 20 mg/l. Brine waters, on the other hand, may contain potassium concentrations in excess of 100 mg/l.

The alkalinity of water is its quantitative capacity to neutralize a strong acid to a designated pH, and is therefore a measure of a gross

property of water. Accurate interpretations in terms of specific substances can be made only when the chemical composition of the sample is known. The alkalinity of most natural surface water is primarily a function of carbonate and bicarbonate content. The alkalinity is taken as an indication of the concentrations of these constitutents. Raw domestic wastewater has an alkalinity only slightly greater than that of the water supply. Properly operating anaerobic digesters typically produce supernatant alkalinities in the range of 2000-4000 mg/l as $CaCO_3$.

METHODS AND MATERIALS

Water samples and associated mosquito larvae were collected from June 1978 through October 1979. Surveillance sites consisted of ground cavities, artificial containers, and storm sewer catch basins. All study sites were located in populated areas of the Lafayette, Indiana vicinity.

Surveillance Site Selection

The selection of surveillance sites was based on accessability, proximity to populated areas, and the potential for continual mosquito breeding. Initially, an exagerated number of sites were considered for study with the thought that all would not prove suitable for long-term monitoring. As sites were found unsuitable, they were eliminated. Some selected sites still did not prove suitable for the entire study period. New sites were added during the 1979 breeding season to yield a total of 20 sites from which suitable data were collected. These sites and their locations are summarized in Table 2.

Surveillance Site Descriptions

Study site 1, a ground cavity, was located west of State Road 526 in the Purdue University Horticulture Park (Figure 1). Surveillance was accomplished during early 1978, but was discontinued due to inaccessibility during wet weather and lack of water retention during dry periods. Study sites 2 and 3 consisted of 2 steel drums located in an open field.

Table 2. Summary of selected study sites.

Site	Habitat Type[a]	Location
1	GC	Horticulture Park, Purdue University
2	AC	Howard Avenue, West Lafayette
3	AC	Howard Avenue, West Lafayette
4	GC	State Road 526, West Lafayette
5	GC	State Road 126, West Lafayette
6	SSCB	Rose Street, West Lafayette
7	SSCB	West Stadium Avenue, West Lafayette
8	GC	Tower Drive, Purdue University
9	AC	State Road 26 West, West Lafayette
10	AC	State Road 26 West, West Lafayette
11	AC	State Road 26 West, West Lafayette
12	AC	State Road 26 West, West Lafayette
13	SSCB	Veterinary School, Purdue University
14	SSCB	Terry Lane, West Lafayette
15	SSCB	Halsey Drive, Purdue University
16	SSCB	South 7th Street, Lafayette
17	SSCB	Bennett Drive, Lafayette
18	SSCB	Lourdes Lane, Lafayette
19	AC	County Road 300 South, Lafayette
20	SSCB	Airport Road, Purdue University

[a]AC = artificial container; GC = ground cavity; SSCB = storm sewer catch basin

Figure 1. Study site 1.

Both sites produced mosquito larvae during the 1978 season, but were destroyed during the winter of 1978-1979. Site 3 is pictured in Figure 2. The drums usually contained from 8-20 cm of water. Constant exposure to the sun resulted in periodic drying. Larvae were generally collected from 7-10 da after a measurable rainfall (1-2 in).

Study site 4 was a roadside drainage ditch (Figure 3). Collections were made from this site during 1978 only. No standing water was found at this site in 1979. Study site 5, also a roadside drainage ditch (Figure 4), was interesting but unpredictable. It produced mosquito larvae after periods of measurable rainfall (1-2 in). Water depths ranged from 3-10 cm.

Study site 6 was monitored from early 1978 to the conclusion of the study. This storm sewer catch basin never became dry and usually contained about 53 cm of water with the water surface 96 cm below street level. Construction was of the poured concrete type, capped by a large steel grate with a curb inlet (Figure 5). The 71 cm top opening expanded to 1 m at the bottom of the sewer. Study site 7 was a 71 cm by 46 cm rectangular storm sewer of brick construction. This sewer was covered by a small steel grate set into the curb (Figure 6). Approximately 20 cm of water was present during most of the summer at a distance of 71 cm below street level.

Study site 8, a 45 cm deep ditch (Figure 7), was created by erosion from drainage water. The substrate of the ditch was primarily silt caused by erosion of the surrounding terrain. This eroding action resulted in a high degree of turbidity. This site was located in a densely wooded area.

Figure 2. Study site 3.

Figure 3. Study site 4.

Figure 4. Study site 5.

Figure 5. Study site 6.

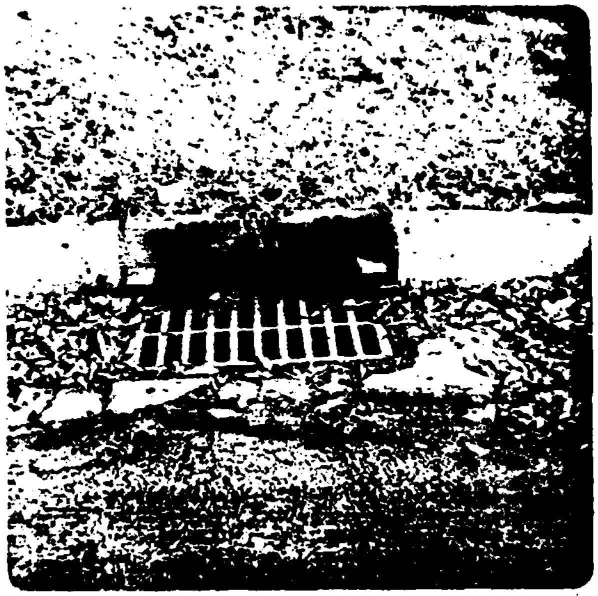


Figure 6. Study site 7.

Figure 7. Study site 8.

Study sites 9, 10, 11, and 12 were located near a large woodlot. Site 9, a 55 gal steel drum (Figure 8), produced mosquito larvae during the summer of 1978 only. While site 10 (Figure 9) was a 30 gal steel drum, sites 11 (Figure 10) and 12 (Figure 11) were 55 gal steel drums. These sites produced mosquito larvae during the entire 2-yr period. All 4 drums were approximately 3/4 filled with earth, debris, and organic materials. Site 10 also contained a number of grass plants. These 4 sites were the most rural of all sites monitored during the study.

Study site 13 was a storm sewer catch basin of poured concrete construction, 61 cm by 61 cm, with a curb inlet and small steel grate (Figure 12). A water depth of 36 cm persisted throughout the breeding season at approximately 1 m below pavement level. This site was important from a potential disease standpoint, as the area immediately surrounding it was utilized as a holding pen for equines.

Study site 14 was a storm sewer catch basin of the poured concrete type, 56 cm by 56 cm, capped by a steel grate with a curb inlet (Figure 13). The site usually contained approximately 15 cm of water with the surface 46 cm below pavement level. This site was continually monitored during the 1979 breeding season. Study site 15 was another storm sewer catch basin of poured concrete construction, with a curb inlet and small steel grate (Figure 14). The water level averaged 18 cm, with the surface 56 cm below pavement level. Monitoring of this site was continuous during the 1979 season. Another storm sewer catch basin, study site 16, was of poured concrete construction, 71 cm in diameter, and capped by a small steel grate with a curb inlet (Figure 15). A constant water depth of 25 cm was maintained at a depth of 86 cm below pavement level.

Figure 8. Study site 9.

Figure 9. Study site 10.

Figure 10. Study site 11.

Figure 11. Study site 12.

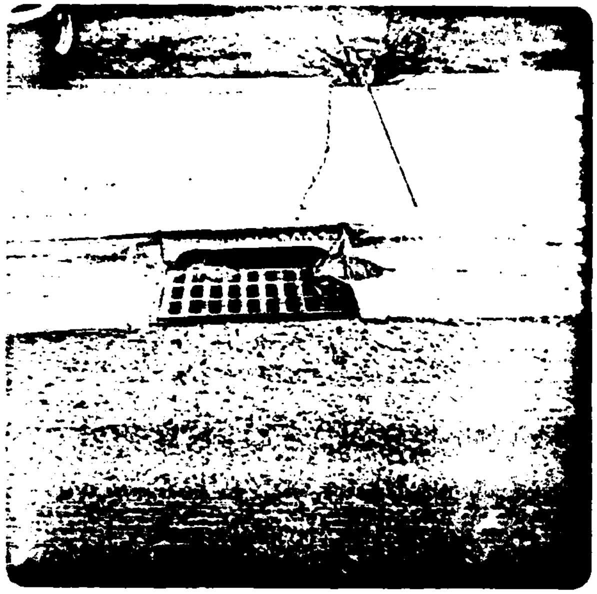


Figure 12. Study site 13.

Figure 13. Study site 14.

Figure 14. Study site 15.

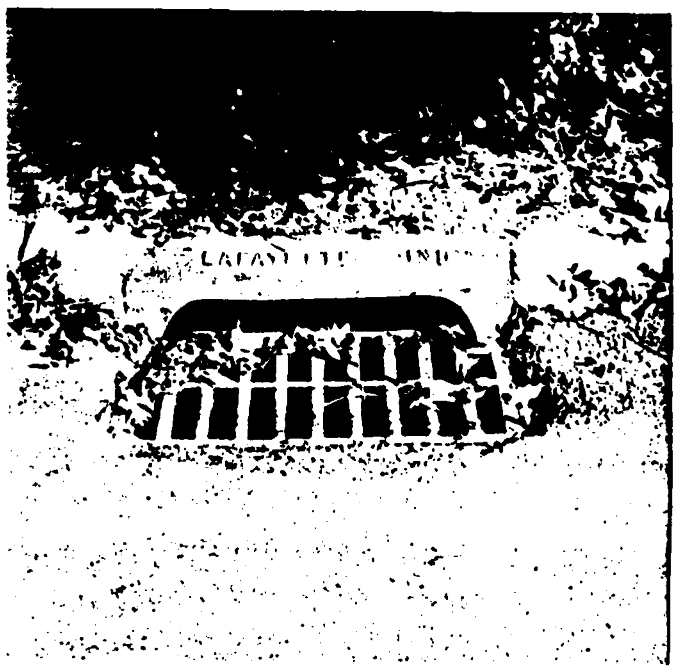


Figure 15. Study site 16.

Mosquito breeding occurred sporadically during the 1979 season, as oil was found on the water surface a number of times.

Study site 17 was a poured concrete storm sewer, 61 cm in diameter and capped by a small steel grate with a curb inlet (Figure 16). The average 10 cm water depth was 86 cm below pavement level. The site was situated near a large wooded ravine. Study site 18, another storm sewer catch basin, was of poured concrete construction, 61 cm in diameter and capped by a small steel grate with a curb inlet (Figure 17). A constant water level of 25 cm was maintained at a distance of 1.5 m below pavement level. This storm sewer was the deepest of all catch basins monitored during the investigation.

Study site 19 was a 5.5 m diameter plastic swimming pool, 46 cm in depth (Figure 18). It was periodically filled with well water which remained in the pool for periods of not less than 10-14 da at a time. The pool was used infrequently, never covered, and chemicals were not added at any time. It was situated in the back corner of a large lot and was surrounded by pine trees.

Study site 20, a storm sewer catch basin, was established during the 1979 breeding season. This site was of poured concrete construction, capped by a small steel grate with a curb inlet (Figure 19). A water depth of 15 cm was maintained at a distance of 66 cm below pavement level. This site was located in close proximity to a large complex of multi-family housing and several athletic fields.

Larval Collection Techniques

Larval mosquito collections were made at weekly intervals from each surveillance site. Larvae were collected, even though insufficient

Figure 16. Study site 17.

Figure 17. Study site 18.

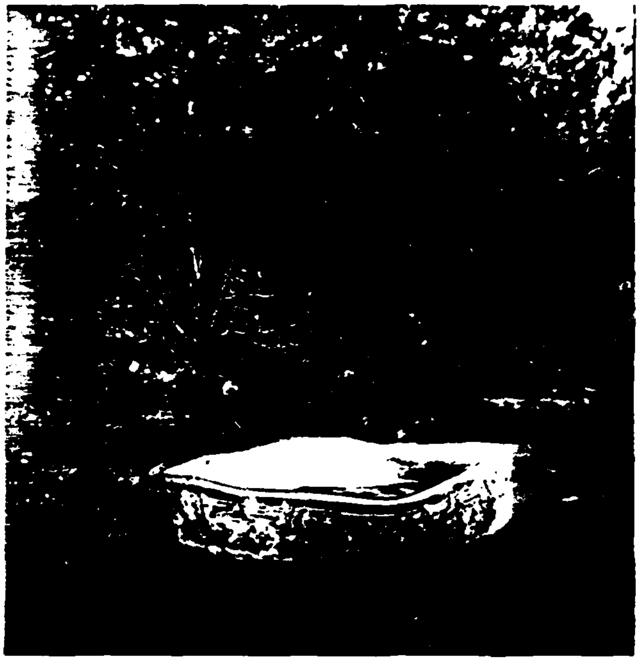


Figure 18. Study site 19.

Figure 19. Study site 20.

amounts of water were availabe for analysis. Larvae were collected with a pint dipper (Figure 20) and transferred to vials containing 70% alcohol. First and second instar larvae were maintained in original breeding water and reared until determinations could be made. A set standard of 5 dips were taken at each surveillance site. The larvae were then transported to the laboratory for identification and logging. "Mosquitoes of Indiana" (Siverly 1972) was utilized as the sole identification key. All larval identifications were performed by the author and confirmed by Dr. Ronald A. Ward, Medical Entomology Project, Smithsonian Institution, Washington, D.C.. The majority of specimens were maintained for future reference. Several specimens of each major species were mounted on slides for comparative purposes. Specimen mounting was accomplished by personnel of the Medical Entomology Project.

On-site Analyses

On-site analysis methods were utilized to measure physical and environmental variables which could be attained in the field. Water temperatures were taken at depths midway between the surface and the bottom of the site so as to avoid inconsistancy. pH was measured with a narrow range colorimeter. The colorimeter chamber was rinsed and then filled with water to be tested. Five drops of phenol red were added and a direct color comparison made. Due to the non-availability of a field pH meter, the results were checked with narrow range pH paper.

Dissolved oxygen was determined using the Hach Kit® titration method. A standard DO bottle was used and carefully filled with the test water. The premeasured reagent packets (Figure 21) were added and the bottle was stoppered and agitated. After the formed flock settled, the

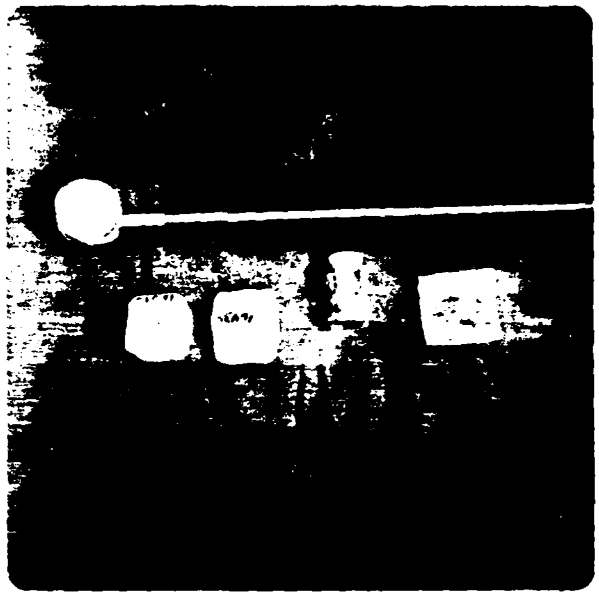


Figure 20. Larval sampling and water collection equipment.

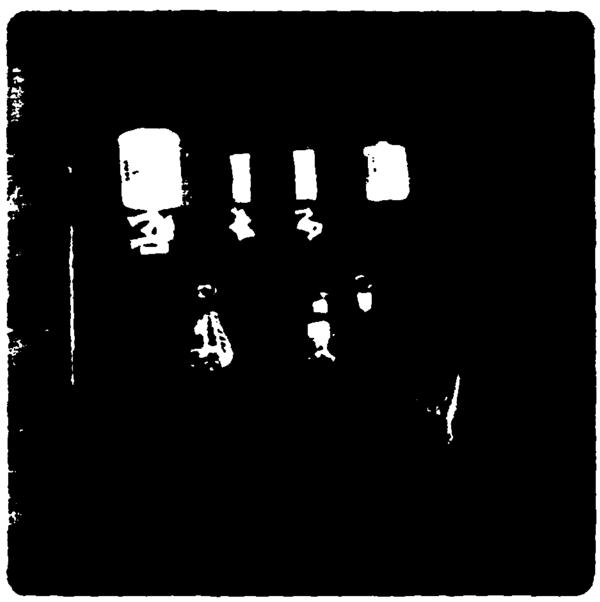


Figure 21. Hach Kit® equipment for measurement of dissolved oxygen in the field.

acid fixer was added and the bottle was again agitated. After the flock disappeared, 5.8 ml of solution was titrated into a clean beaker. Titrant was then added, a drop at a time, until the brown solution became clear. The actual DO value was then interpreted as 1 mg/l, per drop of titrant added.

Environmental observations were made and noted during initial sampling operations. The environmental factors considered included the amount of vegetative cover, exposure to the sun, water movement, aquatic vegetation, and associated aquatic fauna. A summary of these environmental factors was assembled for each study site, and from then on, only unique observations of physical changes were documented.

Water Sample Collection Methods

Water samples for chemical analyses were collected in conjunction with larval surveillance. Collections were made at weekly intervals throughout the breeding season. When insufficient amounts of water were available for collection, applicable on-site analyses were still accomplished if possible.

The water samples were collected with a pint dipper and transferred to 1000 ml polyethylene water collection cubits (Figure 20). The water samples were filtered through a 60-mesh wire screen to remove mosquito larvae and debris. A second water sample of 100 ml was collected in the same manner and transferred to a clean glass jar. This sample was preserved with 0.2 ml concentrate sulfuric acid (H_2SO_4) and submitted for analysis of COD. All other laboratory analyses were performed on the 1000 ml unpreserved sample.

Water samples were identified by site number, date, collector, and sample submission number. An accompanying submission form was accomplished, indicating the results of on-site analyses and the laboratory tests to be performed. The samples were packaged and mailed to the United States Air Force Occupational and Environmental Health Laboratory, Brooks Air Force Base, San Antonio, Texas, for analyses.

Laboratory Analyses of Variables

Analyses of variables selected for this study were accomplished in accordance with accepted standard procedures for the examination of water and wastewater (American Public Health Association 1981). The procedures utilized are intended for the physical and chemical examination of natural waters in the absence of gross pollution. None of the surveillance sites selected for this study displayed any evidence of wastewater or similar gross pollution.

Chemical oxygen demand determinations were accomplished by reflux with known amounts of potassium dichromate and H_2SO_4. The excess dichromate was titrated with ferrous ammonium sulfate. The amount of oxidizable organic matter, measured as oxygen equivalent, was then proportional to the potassium dichromate consumed. After calculation, the resultant COD values were reported in mg/l.

Specific conductance was measured using an Rd Solu Bridge conductivity meter at 25°C. The results were reported in micromhos/cm. Calcium determinations were accomplished using the permanganate tetrimetric method. Precipitated calcium oxalate was redissolved in H_2SO_4 and titrated with permanganate. The amount of permanganate required to oxidize the oxalate is proportional to the amount of calcium. Results were

reported in mg/l. Magnesium levels were determined in conjunction with the calcium analysis. Oxalate and ammonium salts were removed from the combined filtrate and washings which remained from the calcium determination. This residue was again washed and filtered. The resultant filtrate was heated and the residue ignited for 30 min periods at 1100°C until a constant weight was maintained. The magnesium level was then determined based on the known residue weight and reported in mg/l.

Total hardness was determined by calculation from the results of the calcium and magnesium analyses. The concentration of the cations was multiplied by the proper factor (calcium = 2.479, magnesium = 4.116) to obtain equivalent $CaCO_3$ concentrations. These concentrations were then summed and reported in mg/l $CaCO_3$.

Sodium and potassium levels were determined by the flame photometric method. Direct-intensity measurements were performed on both calibration standards and samples in order to obtain a reliable average for each solution. Sodium and potassium concentrations were determined by consulting a calibration curve. The results of each analysis were reported in mg/l.

Analysis for sulfate was done using the gravimetric method in which the residue was ignited. Cation interference and silica were removed from the sample and barium sulfate was precipitated from the combined filtrate and washings. The precipitate was then washed and filtered to remove the chloride. The remaining precipitate and filter were ignited at 800°C, cooled, and the residue weighed. Sulfate values were reported in mg/l.

Chloride determinations were made using the argentometric method. Samples were adjusted to a pH range of from 7.0-10.0, if necessary. Potassium chromate indicator solution was added to the sample and titration to a pinkish-yellow endpoint was done utilizing standard silver nitrate titrant. The silver nitrate titrant was then standardized and a reagent blank value established. From these known values, the chloride determination was calculated and reported in mg/l.

Alkalinity determinations were made using indicator methods. These methods proved adequate due to the relatively high alkalinity concentrations observed in the initial samples. Phenolphthalein indicator was added to the sample and titrated with 0.02 normal standard acid until a coloration endpoint of pH 8.3 was reached. Methyl orange indicator was then added to this same solution and again titrated with 0.02 normal standard acid to the proper equivalence point. Total alkalinity was then determined and reported as mg/l $CaCO_3$. The carbonate and bicarbonate alkalinity relationships were then determined. These values were based on the results of the phenolphthalein indicator portion of the total alkalinity determination. Both carbonate and bicarbonate alkalinity were also reported in mg/l $CaCO_3$.

Data Analyses Methods

Larval collection data was analyzed on the basis of overall, habitat type, and individual species data. The grouped data is reported on a monthly basis by average number of specimens per collection and by the % make-up of monthly species collections. Indices of association were calculated to determine the degree of association or disassociation between species collected from the same habitat.

Statistical analyses of environmental and water chemistry data were done by multiple linear regression. The program was written so that it would determine the effect of each independent variable when removed, 1 at a time, from the overall equation. The program provided a rank order by variable significance and correlation coefficients relating to variable interaction.

RESULTS AND DISCUSSION

Two seperate data sets were collected and analyzed during the course of this research. The analysis of larval breeding collections is presented first and later correlated with the water chemistry data. A thorough understanding of area mosquito breeding by habitat type, as well as species seasonal breeding progression, is essential before breeding site water chemistry data correlations can be made.

Overall Larval Mosquito Surveillance

A total of 11 mosquito species were collected from the 3 habitat types during the 2-yr study period. Table 3 lists the relative numerical importance of the mosquito species collected in the Lafayette, Indiana area. The most abundant species collected was *Cx. restuans* (28.6%), followed by *Cx. salinarius* (25.4%), *Ae. triseriatus* (23.1%), and *Cx. pipiens* (18.9%). The remaining 7 species were collected much less frequently.

The overall % make-up of monthly larval collection totals, as well as the average number of specimens collected are presented in Table 4. The majority of specimens were collected during July (27.2%), August (29.7%), and September (21.7%), the months of greatest mosquito-borne disease activity in the Midwest. The relative abundance of proven or suspected disease vector species was highest during the August/September period, indicating an existing potential for disease transmission.

Table 3. Relative numerical importance of larval mosquitoes collected at Lafayette/West Lafayette, Indiana.

Species	Total Collected 1978-1979	Yearly Totals 1978	Yearly Totals 1979	Yearly % 1978	Yearly % 1979	% of Total 1978-1979
Aedes triseriatus	1119	50	1069	4.47	95.53	23.05
Aedes vexans	72	45	27	62.50	37.50	1.48
Anopheles punctipennis	70	50	20	71.43	28.57	1.44
Culex pipiens	915	517	398	56.50	43.50	18.85
Culex restuans	1390	465	925	33.45	66.55	28.63
Culex salinarius	1233	680	553	55.15	44.85	25.40
Culex territans	35	10	25	28.57	71.43	0.72
Culiseta inornata	3	3	0	100.00	0	0.06
Orthopodomyia signifera	16	0	16	0	100.00	0.33
Psorophora columbiae	1	1	0	100.00	0	0.02
Psorophora howardii	1	1	0	100.00	0	0.02
Totals	4855	1822	3033	37.53	62.47	100.00

Table 4. Larval composition of all monitored study sites.

Species	%						
	May	Jun	Jul	Aug	Sep	Oct	Total
Aedes triseriatus	14.24 (22.50)[a]	0 (0)	38.73 (46.55)	28.60 (45.89)	14.15 (18.68)	0 (0)	23.05 (37.30)
Aedes vexans	0 (0)	7.55 (15.00)	0.98 (4.33)	0 (0)	0 (0)	11.29 (7.00)	1.48 (9.00)
Anopheles punctipennis	0 (0)	0 (0)	3.33 (14.67)	1.46 (10.50)	0.48 (5.00)	0 (0)	1.44 (11.67)
Culex pipiens	0 (0)	8.39 (25.00)	11.27 (14.90)	25.76 (16.22)	31.62 (17.47)	8.87 (5.50)	18.85 (16.34)
Culex restuans	85.76 (54.00)	74.33 (37.08)	31.39 (29.64)	16.55 (15.87)	2.09 (4.40)	0 (0)	28.63 (27.26)
Culex salinarius	0 (0)	8.89 (13.00)	13.69 (18.10)	25.76 (21.88)	50.14 (25.19)	79.84 (24.75)	25.40 (22.02)
Culex territans	0 (0)	0 (0)	0.61 (2.67)	1.87 (5.40)	0 (0)	0 (0)	0.72 (4.38)
Culiseta inornata	0 (0)	0.50 (3.00)	0 (0)	0 (0)	0 (0)	0 (0)	0.06 (3.00)
Orthopodomyia signifera	0 (0)	0 (0)	0 (0)	0 (0)	1.52 (8.00)	0 (0)	0.33 (8.00)
Psorophora columbiae	0 (0)	0.17 (1.00)	0 (0)	0 (0)	0 (0)	0 (0)	0.02 (1.00)
Psorophora howardii	0 (0)	0.17 (1.00)	0 (0)	0 (0)	0 (0)	0 (0)	0.02 (1.00)
Totals	6.51 (45.00)	12.28 (42.64)	27.23 (44.07)	29.74 (43.76)	21.69 (35.10)	2.55 (17.86)	100.00 (40.12)

[a]() indicate average number of specimens per collection

From the data presented in Table 4, an overall seasonal breeding progression is evident. Early season dominance by Cx. restuans, eventually gave way to building populations of Cx. pipiens and Cx. salinarius. Competition between the later 2 species was indicated throughout the season. An index of association was calculated to determine the degree of association or disassociation between 2 mosquito species occurring at the same location. The chosen index of association is represented by the formula,

$$I = 2\left[\frac{Ji}{A + B} - 0.5\right]$$

where Ji is the number of both species in samples where they occur together, and A and B are the total number of individuals of the 2 species in all samples. The numerical value of I lies between +1 and -1, indicating either a strong degree of association, or a strong degree of disassociation. Values closer to 0 indicate the lack of either type of association. The stronger the degree of positive association, the greater the competition factor among species.

An index of association was calculated for each of 3 major species pairs based on overall surveillance data. The species pairs and corresponding indices of association are as follows: Cx. pipiens/Cx. salinarius, I = +0.26; Cx. pipiens/Cx. restuans, I = -0.28; and Cx. salinarius/Cx. restuans, I = -0.53. The calculated I values closely parallel the overall seasonal breeding progressions of the pairs involved. The I of +0.26 for the Cx. pipiens/Cx. salinarius pair indicates a degree of association and a related competition factor. As the population of 1 species increases, the other decreases and vice versa. On the other hand, the I values of the remaining 2 pairs were both negative, indicating a

disassociation and subsequent lack of competition.

Identified breeding locations and habitat types are equally as important as species presence. The average number of larval specimens per collection and the % of monthly collection totals for each surveillance site type are contained in Table 5. The artificial container breeding sites provided both the highest average number of specimens per collection (42.9) and the highest % of total specimens (43.4%). The storm sewer catch basin breeding sites ranked second in both categories with 39.7 and 42.5%, respectively. Mosquito breeding occurred in these 2 site types from May into October. On the other hand, decreased rainfall and the effects of drying in late summer and early fall had more impact on ground cavity breeding sites. These sites contained mosquito larvae only from May through August. The lack of late season breeding resulted in a lower % of total specimens from the ground cavity sites (14.1%), although the average number of specimens per collection remained relatively high at 34.3.

All 3 habitat types can be considered major contributors to the local mosquito problem. A closer evaluation of breeding in each habitat type is in order to more fully understand seasonal breeding progressions and species interactions.

Habitat Type and Individual Species Surveillance

A total of 9 storm sewer catch basin study sites were monitored over the 2-yr surveillance period. Mosquito species routinely collected from area catch basins included *Cx. pipiens*, *Cx. restuans*, and *Cx. salinarius*. *Ae. vexans* and *Cx. territans* were collected on a single occasion only. Table 6 summerizes the composition of monthly larval collection totals

Table 5. Larval composition of monitored habitat types.

Habitat Type	%						
	May	Jun	Jul	Aug	Sep	Oct	Total
Storm Sewer Catch Basin	62.03 (65.00)[a]	60.57 (45.38)	17.78 (39.17)	40.51 (36.50)	55.94 (42.07)	79.03 (19.60)	42.51 (39.69)
Artificial Container	14.24 (22.50)	21.14 (42.00)	60.67 (57.29)	44.46 (53.50)	44.06 (29.00)	20.97 (13.00)	43.36 (42.96)
Ground Cavity	23.73 (37.50)	18.29 (36.00)	21.55 (28.50)	15.03 (43.60)	0 (0)	0 (0)	14.13 (34.30)
Total	6.51 (45.00)	12.28 (42.64)	27.23 (44.07)	29.74 (43.76)	21.69 (35.10)	2.55 (17.86)	100.00 (40.12)

[a]() indicate average number of specimens per collection

Table 6. Larval composition of all monitored storm sewer catch basin study sites.

Species	%						
	May	Jun	Jul	Aug	Sep	Oct	Total
Aedes vexans	0 (0)[a]	0 (0)	0 (0)	0 (0)	0 (0)	14.29 (7.00)	0.68 (7.00)
Culex pipiens	0 (0)	13.77 (25.00)	8.94 (4.20)	36.99 (16.62)	46.35 (24.82)	6.12 (6.00)	27.42 (17.69)
Culex restuans	100.00 (65.00)	82.37 (37.38)	33.19 (15.60)	23.80 (12.64)	3.73 (4.40)	0 (0)	35.51 (22.91)
Culex salinarius	0 (0)	3.86 (7.00)	57.87 (27.20)	38.87 (20.64)	49.92 (29.40)	79.59 (26.00)	36.29 (24.16)
Culex territans	0 (0)	0 (0)	0 (0)	0.34 (1.00)	0 (0)	0 (0)	0.10 (1.00)
Total	9.49 (65.00)	17.59 (45.38)	11.39 (39.17)	28.29 (36.50)	28.54 (42.07)	4.74 (19.60)	100.00 (39.69)

[a]() indicate average number of specimens per collection

for all storm sewer catch basin study sites.

The most abundant storm sewer breeder was Cx. salinarius (36.3%), followed by Cx. restuans (35.5%), and Cx. pipiens (27.4%). The highest catch basin breeding levels occurred during the months of August and September in almost equal levels. Larger individual collections were noted in the spring of the year, leveling off during mid-summer, and rapidly decreasing in the fall.

Cx. restuans was the most abundant species collected during this study (28.6%). As indicated in Table 7, it was collected from all 3 habitat types and was present from May through September in storm sewer catch basins. Cx. restuans was most abundant early in the breeding season and then decreased in relative abundance through September. The average early season number of specimens per collection was higher for Cx. restuans than for any other species.

Cx. salinarius, the second most abundant species (25.4%), was consistently taken from storm sewer catch basins and artificial containers from June through October. A single collection was made from a ground cavity site during August. As indicated in Table 8, Cx. salinarius was most abundant during August and September, with the majority of specimens collected from catch basin sites (60.8%).

A steady seasonal increase in Cx. salinarius population levels was observed from both storm sewer catch basin and artificial container breeding sites. With respect to these habitats, when the population of 1 site type increased, the other decreased and vice versa. This trend continued throughout the mosquito breeding season and effectively resulted in the maintenance of a relatively high Cx. salinarius population level at all times. It was the dominant species in early summer, but by

Table 7. *Culex restuans* larval composition of monitored habitat types.

Habitat Type	%						
	May	Jun	Jul	Aug	Sep	Oct	Total
Storm Sewer Catch Basin	72.22 (65.00)[a]	67.19 (37.38)	18.80 (15.60)	58.40 (12.64)	100.00 (4.40)	0 (0)	52.73 (22.91)
Artificial Container	0 (0)	19.78 (29.33)	28.19 (19.50)	15.55 (18.50)	0 (0)	0 (0)	17.41 (22.00)
Ground Cavity	27.78 (37.50)	13.03 (58.00)	53.01 (73.33)	26.05 (31.00)	0 (0)	0 (0)	29.86 (51.88)
Total	19.43 (54.00)	32.01 (37.08)	29.86 (29.64)	17.12 (15.87)	1.58 (4.40)	0 (0)	100.00 (27.26)

[a]() indicate average number of specimens per collection

Table 8. Culex salinarius larval composition of monitored habitat types.

Habitat Type	%						
	May	Jun	Jul	Aug	Sep	Oct	Total
Storm Sewer Catch Basin	0 (0)[a]	26.92 (7.00)	75.14 (27.20)	61.02 (20.64)	55.58 (29.40)	78.79 (26.00)	60.75 (24.16)
Artificial Container	0 (0)	73.08 (19.00)	24.86 (9.00)	38.44 (28.60)	44.42 (21.36)	21.21 (21.00)	39.09 (20.08)
Ground Cavity	0 (0)	0 (0)	0 (0)	0.54 (2.00)	0 (0)	0 (0)	0.16 (2.00)
Total	0 (0)	4.22 (13.00)	14.68 (18.10)	30.17 (21.88)	42.90 (25.19)	8.03 (24.75)	100.00 (22.02)

[a]() indicate average number of specimens per collection

late summer only slightly more abundant than Cx. pipiens, the fourth most abundant species (18.9%).

The majority of Cx. pipiens specimens were collected from storm sewer catch basins as indicated in Table 9. The species was most abundant during August and September. By early fall, Cx. pipiens population levels fell off sharply and Cx. salinarius dominated until the end of the breeding season.

The indices of association for the major storm sewer catch basin species were calculated in support of competition factors indicated by seasonal breeding progressions. The species pairs and corresponding indices of association are as follows: Cx. pipiens/Cx. salinarius, I = +0.31; Cx. pipiens/Cx. restuans, I = -0.11 and; Cx. salinarius/Cx. restuans, I = -0.14. The calculated I values for the catch basin breeders are similar to those for the overall data, but show a stronger degree of association between Cx. pipiens and Cx. salinarius.

Eight of the 9 individual storm sewer catch basin sites contained mosquito larvae at 1 time or another during the course of the study. The sites from which several species were collected fit the overall seasonal breeding progression for this site type. Table 10 provides a comparative summary of species breeding at each study site within the storm sewer catch basin habitat type. All sites were monitored several times each month, but obviously collected only when the site contained water. During months when no collections were made, the site was dry. The most unusual storm sewer catch basin was site 18. It contained water continually but never produced mosquito larvae. This particular catch basin was by far the deepest of those monitored.

Table 9. _Culex pipiens_ larval composition of monitored habitat types.

Habitat Type	%						
	May	Jun	Jul	Aug	Sep	Oct	Total
Storm Sewer Catch Basin	0 (0)[a]	100.00 (25.00)	14.09 (4.20)	57.91 (16.62)	82.23 (24.82)	54.55 (6.00)	61.86 (17.69)
Artificial Container	0 (0)	0 (0)	85.91 (25.60)	12.87 (6.00)	17.77 (7.38)	45.45 (5.00)	26.23 (10.91)
Ground Cavity	0 (0)	0 (0)	0 (0)	29.22 (54.50)	0 (0)	0 (0)	11.91 (54.50)
Total	0 (0)	5.47 (25.00)	16.28 (14.90)	40.77 (16.22)	36.28 (17.47)	1.20 (5.50)	100.00 (16.34)

[a]() indicate average number of specimens per collection

Table 10. Larval composition of the 9 individual study sites of the storm sewer catch basin habitat type.

Species	% by Site								
	6	7	13	14	15	16	17	18	20
Aedes vexans	0	3	0	0	0	0	0	0	0
Culex pipiens	21	28	47	0	0	31	0	0	100
Culex restuans	23	43	1	90	0	45	95	0	0
Culex salinarius	55	26	52	10	100	24	5	0	0
Culex territans	1	0	0	0	0	0	0	0	0

The 7 artificial container study sites monitored yielded 7 species of mosquito larvae. In order of abundance they included, Ae. triseriatus (53.2%), Cx. salinarius (22.9%), Cx. restuans (11.5%), Cx. pipiens (11.4%), Or. signifera (0.8%), An. punctipennis (0.2%), and Cx. territans (0.04%). The % make-up of monthly collection totals for these artificial container species is contained in Table 11.

The highest breeding levels occurred during July and August, with both months dominated by Ae. triseriatus, the third most abundant species overall (23.1%). It was the only species collected from monitored artificial container sites during the month of May. The monthly collection data for Ae. triseriatus are summarized in Table 12. All specimens were collected from 55 gal steel drums located near a densely wooded area. Breeding occurred from May through September with a break in June, due to drying and low water levels in each of the sites.

Ae. triseriatus population levels seemed little effected by the presence of other species. Cx. restuans displayed similar artificial container monthly population levels as were found in storm sewer catch basin collections. Early season levels were high, gradually decreasing through August, and finally tailing off in September. Populations of Cx. salinarius and Cx. pipiens showed definite competition throughout the breeding season. Cx. salinarius emerged during the month of June. Upon the emergence of Cx. pipiens during July, the Cx. salinarius population fell off considerably. The reverse occurred during August, when Cx. salinarius dominated and Cx. pipiens declined sharply. Cx. salinarius continued to dominate through the end of the breeding season.

Table 11. Larval composition of all monitored artificial container study sites.

	%						
	May	Jun	Jul	Aug	Sep	Oct	Total
Aedes triseriatus	100.00 (22.50)[a]	0 (0)	63.84 (46.55)	64.33 (45.89)	32.11 (18.63)	0 (0)	53.16 (37.30)
Anopheles punctipennis	0 (0)	0 (0)	0 (0)	0 (0)	1.08 (5.00)	0 (0)	0.24 (5.00)
Culex pipiens	0 (0)	0 (0)	15.96 (25.60)	7.48 (6.00)	12.71 (7.38)	19.23 (5.00)	11.40 (10.91)
Culex restuans	0 (0)	69.84 (29.33)	14.59 (19.50)	5.76 (18.50)	0 (0)	0 (0)	11.50 (22.00)
Culex salinarius	0 (0)	30.16 (19.00)	5.61 (9.00)	22.27 (28.60)	50.65 (21.36)	80.77 (21.00)	22.90 (20.08)
Culex territans	0 (0)	0 (0)	0 (0)	0.16 (1.00)	0 (0)	0 (0)	0.04 (1.00)
Orthopodomyia signifera	0 (0)	0 (0)	0 (0)	0 (0)	3.45 (8.00)	0 (0)	0.76 (8.00)
Total	2.14 (22.50)	5.99 (42.00)	38.10 (57.29)	30.50 (53.50)	22.04 (29.00)	1.23 (13.00)	100.00 (42.96)

[a]() indicate average number of specimens per collection

Table 12. *Aedes triseriatus* larval composition of monitored habitat types.

Habitat Type	%						
	May	Jun	Jul	Aug	Sep	Oct	Total
Storm Sewer Catch Basin	0 (0)[a]	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Artificial Container	100.00 (22.50)	0 (0)	100.00 (45.55)	100.00 (45.89)	100.00 (18.63)	0 (0)	100.00 (37.30)
Ground Cavity	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Total	4.02 (22.50)	0 (0)	45.76 (45.55)	36.91 (45.89)	13.31 (18.63)	0 (0)	100.00 (37.30)

[a]() indicate average number of specimens per collection

The indices of association, calculated for the major artificial container species, are as follows: Cx. pipiens/Cx. salinarius, I = +0.26; Cx. pipiens/Cx. restuans, I = +0.03; Cx. pipiens/Ae. triseriatus, I = -0.52; Cx. restuans/Cx. salinarius, I = -0.57; Cx. salinarius/Ae. triseriatus, I = -0.71 and; Cx. restuans/Ae. triseriatus, I = -0.89. Again, the Cx. pipiens/Cx. salinarius pair shows a positive index of association. The Cx. pipiens/Cx. restuans pair shows a very slight degree of positive association, more than likely created by the smaller breeding area of the artificial container habitats.

A known artificial container breeder, Or. signifera, was taken from a 55 gal steel drum on 2 occasions during early September. The site was located near a wooded area quite suitable for breeding of this species. Table 13 offers a comparative summary of species breeding at each artificial container study site.

Collection site 19 was the only artificial container which did not produce mosquito larvae. This small swimming pool contained water most of the breeding season. It is thought that disturbances by feral animals, as well as periodic semi-drying, may have influenced larval development by causing fluctuations in the water level.

All 4 monitored ground cavity sites contained larvae at 1 time or another during the surveillance period. The dominate ground cavity species was Cx. restuans, accounting for 60.5% of the total specimens collected. The remaining ground cavity species included Cx. pipiens (15.9%), An. punctipennis (9.5%), Ae. vexans (8.5%). Cx. territans (4.7%), Cu. inornata (0.4%), Cx. salinarius (0.3%), and Ps. columbiae and Ps. howardii (0.2%). This collection data is summarized in Table 14.

Table 13. Larval composition of the 7 individual study sites of the artificial container habitat type.

Species	% by Site						
	2	3	9	10	11	12	19
Aedes _triseriatus_	0	0	0	16	94	84	0
Anopheles _punctipennis_	0	0	0	0	1	0	0
Culex _pipiens_	15	5	0	32	1	7	0
Culex _restuans_	45	48	51	9	0	0	0
Culex _salinarius_	40	47	49	40	4	9	0
Culex _territans_	0	0	0	0	1	0	0
Orthopodomyia _signifera_	0	0	0	3	0	0	0

Table 14. Larval composition of all monitored ground cavity study sites.

Species	%						
	May	Jun	Jul	Aug	Sep	Oct	Total
Aedes vexans	0 (0)[a]	41.66 (15.00)	4.56 (4.33)	0 (0)	0 (0)	0 (0)	8.45 (9.67)
Anopheles punctipennis	0 (0)	0 (0)	15.44 (14.67)	0 (0)	0 (0)	0 (0)	9.47 (13.00)
Culex pipiens	0 (0)	0 (0)	0 (0)	50.00 (54.50)	0 (0)	0 (0)	15.89 (54.50)
Culex restuans	100.00 (37.50)	53.70 (58.00)	77.19 (73.33)	28.44 (31.00)	0 (0)	0 (0)	60.50 (51.88)
Culex salinarius	0 (0)	0 (0)	0 (0)	0.92 (2.00)	0 (0)	0 (0)	0.29 (2.00)
Culex territans	0 (0)	0 (0)	2.81 (2.67)	11.01 (12.00)	0 (0)	0 (0)	4.66 (6.40)
Culiseta inornata	0 (0)	2.78 (3.00)	0 (0)	0 (0)	0 (0)	0 (0)	0.44 (3.00)
Psorophora columbiae	0 (0)	0.93 (1.00)	0 (0)	0 (0)	0 (0)	0 (0)	0.15 (1.00)
Psorophora howardii	0 (0)	0.93 (1.00)	0 (0)	0 (0)	0 (0)	0 (0)	0.15 (1.00)
Total	10.93 (37.50)	15.74 (36.00)	41.54 (28.50)	31.79 (43.60)	0 (0)	0 (0)	100.00 (34.30)

[a]() indicate average number of specimens per collection

An. punctipennis was found most frequently in ground cavities. A single collection was taken from an artificial container in September. Ae. vexans was also primarily collected from ground cavities, with a single collection made from a storm sewer catch basin in October. Cx. territans was collected from all three habitat types, with the majority of specimens taken from ground cavity sites during July and August. Cu. inornata, Ps. columbiae, and Ps. howardii were each taken from the same ground cavity site during June. This single collection was the only instance in which these species were found.

The major problem encountered from ground cavity breeding sites was periodic drying. Significant amounts of rainfall (1-2 in) were necessary to maintain sufficient water levels for breeding. When breeding did occur, the average number of larvae per collection was relatively high. The monthly population levels of Cx. restuans were again predictable. High early season levels gradually decreased throughout the season and drastically fell off at the end of August. The reason for the earlier season ceasation of breeding was directly attributed to a lack of rainfall. All ground cavity sites were dry by the end of August.

The competition factor among ground cavity species was virtually nonexistent. In contrast to artificial containers and storm sewer catch basins, the surface area of ground cavity sites was considerably larger. This increased area allowed for the coexistence of several species at one time, without placing them in direct competition. Pockets of several species were often found breeding within the same ground cavity. The calculation of indices of association was not feasible.

The most prolific ground cavity sites were 5 and 8. They both contained water periodically from May through August. Though monitored continually, sites 1 and 4 dried rapidly, even after heavy rains. Table 15 provides a comparative summary of species breeding at each site of this habitat type.

Tables 1A through 5A contain individual species collection data for the entire period of study, as well as an association chart, showing the number of times each species was collected by itself or with another species. With this background of species presence and breeding progression in mind, an analysis and correlation of environmental and water chemistry data is in order.

Environmental and Water Chemistry Variable Ranges and Means

The selected environmental and water chemistry variables measured are reported on the basis of overall, habitat type, and individual species data. A variety of data analyses and statistical methods were used to analyze the data. In all cases, the variable ranges and means are reported and compared. In order to establish a baseline for variable values, all collected data was grouped and the ranges and means calculated for each selected variable (Table 16). This data includes all water collections made from all habitat types.

The ranges and means for these same variables were calculated for each of the 3 habitat types, and also for the 4 major study species (Table 16). Included with the habitat data are the ranges and means of all water chemistry data collected from monitored sites on occasions when no larvae were present. All of the reported non-breeding data were collected from storm sewer catch basin sites. The means of the selected

Table 15. Larval composition of the 4 individual study sites of the ground cavity habitat type.

Species	% by Site			
	1	4	5	8
<u>Aedes vexans</u>	0	96	2	3
<u>Anopheles punctipennis</u>	84	0	30	0
<u>Culex pipiens</u>	0	0	0	23
<u>Culex restuans</u>	0	0	44	74
<u>Culex salinarius</u>	0	0	2	0
<u>Culex territans</u>	16	0	20	0
<u>Culiseta inornata</u>	0	0	2	0
<u>Psorophora columbiae</u>	0	2	0	0
<u>Psorophora howardii</u>	0	2	0	0

Table 16. Ranges and means of selected variables for overall, habitat type, non-breeding, and individual species collection data.

Variables	Overall Data
Water Temperature (°C)	13-30 (21.93)[a]
pH (units)	6.4-8.6 (7.61)
Dissolved Oxygen (mg/l)	0-15 (3.28)
Calcium (mg/l)	0.8-145.8 (35.33)
Magnesium (mg/l)	0.8-70.4 (12.42)
Chloride (mg/l)	0.5-3900 (262.65)
Specific Conductance (micromhos/cm)	0.15-12.8 (2.03)
Sulfate (mg/l)	1-2250 (212.29)
Total Alkalinity (mg/l)	46-2903 (605.78)
Carbonate Alkalinity (mg/l)	0-1634 (121.16)
Bicarbonate Alkalinity (mg/l)	46-2257 (484.36)
Total Hardness (mg/l)	6-654 (141.43)
Chemical Oxygen Demand (mg/l)	23-1500 (196.06)
Potassium (mg/l)	2.2-5400 (542.63)
Sodium (mg/l)	2.3-145.1 (36.35)

[a]() indicate mean variable values

Table 16, cont.

Variables	Artificial Container	Ground Cavity
Water Temperature (°C)	13-30 (23.19)	16-30 (20.67)
pH (units)	6.4-8.6 (8.12)	6.8-8.2 (7.38)
Dissolved Oxygen (mg/l)	3-15 (5.06)	0-9 (4.17)
Calcium (mg/l)	0.8-117.2 (22.77)	10.9-145.8 (63.62)
Magnesium (mg/l)	0.8-46.9 (11.06)	7.4-70.4 (31.52)
Chloride (mg/l)	4-3900 (600.77)	8-200 (46.08)
Specific Conductance (micromhos/cm)	0.17-12.8 (4.13)	0.25-5.0 (0.99)
Sulfate (mg/l)	12-2250 (450.48)	8-600 (94.00)
Total Alkalinity (mg/l)	46-2903 (1145.90)	108-1465 (364.83)
Carbonate Alkalinity (mg/l)	0-1634 (272.00)	0-776 (64.67)
Bicarbonate Alkalinity (mg/l)	46-2257 (873.26)	108-689 (300.17)
Total Hardness (mg/l)	6-342 (219.16)	75-654 (291.33)
Chemical Oxygen Demand (mg/l)	28-1500 (301.95)	23-340 (102.80)
Potassium (mg/l)	9.1-5400 (1322.88)	9.1-25.8 (15.08)
Sodium (mg/l)	3.8-145.1 (58.02)	2.3-17.8 (5.93)

Table 16, cont.

Variables	Storm Sewer Catch Basin	Non-breeding
Water Temperature (°C)	14-29 (21.27)	14-24 (18.83)
pH (units)	6.4-8.4 (6.93)	6.4-7.6 (6.94)
Dissolved Oxygen (mg/l)	0-4 (1.27)	0-6 (1.34)
Calcium (mg/l)	2.9-97.4 (36.85)	11.7-102.5 (32.84)
Magnesium (mg/l)	1-43.2 (6.74)	1.5-25.8 (6.03)
Chloride (mg/l)	0.5-200 (23.77)	4-140 (18.12)
Specific Conductance (micromhos/cm)	0.15-1.90 (.445)	0.11-0.70 (.284)
Sulfate (mg/l)	1-400 (31.55)	3-70 (17.56)
Total Alkalinity (mg/l)	52-1118 (186.00)	37-370 (129.09)
Carbonate Alkalinity (mg/l)	0 (0)	0 (0)
Bicarbonate Alkalinity (mg/l)	52-1118 (186.00)	37-370 (129.09)
Total Hardness (mg/l)	12-387 (126.33)	20-324 (106.83)
Chemical Oxygen Demand (mg/l)	34-635 (122.53)	45-560 (136.00)
Potassium (mg/l)	2.2-36.8 (11.10)	0.7-35.5 (6.61)
Sodium (mg/l)	2.8-102.8 (14.42)	0.7-28.8 (7.25)

Table 16, cont.

Variables	*Aedes triseriatus*	*Culex pipiens*
Water Temperature (°C)	13-27 (22.42)	18-30 (23.04)
pH (units)	7.9-8.4 (8.28)	6.4-8.6 (7.39)
Dissolved Oxygen (mg/l)	3-12 (5.26)	0-12 (2.86)
Calcium (mg/l)	0.8-117.2 (23.45)	2.9-56 (24.66)
Magnesium (mg/l)	0.8-25.6 (10.34)	1-46.9 (9.31)
Chloride (mg/l)	12-3900 (560.00)	1-3900 (467.11)
Specific Conductance (micromhos/cm)	0.24-7.50 (3.74)	0.15-14.0 (2.69)
Sulfate (mg/l)	27-1450 (358.63)	1-2250 (349.43)
Total Alkalinity (mg/l)	120-2903 (1192.74)	50-2903 (661.20)
Carbonate Alkalinity (mg/l)	0-1634 (302.95)	0-1634 (135.60)
Bicarbonate Alkalinity (mg/l)	120-2152 (889.79)	50-2257 (527.97)
Total Hardness (mg/l)	6-342 (94.58)	20-290 (107.46)
Chemical Oxygen Demand (mg/l)	155-343 (299.91)	34-1500 (193.50)
Potassium (mg/l)	9.1-3000 (1137.42)	2.7-5400 (1012.49)
Sodium (mg/l)	3.8-145.1 (58.35)	3.1-145.1 (49.09)

Table 16, cont.

Variables	*Culex restuans*	*Culex salinarius*
Water Temperature (°C)	14-30 (21.90)	16-30 (22.71)
pH (units)	6.4-8.4 (7.08)	6.4-8.6 (7.42)
Dissolved Oxygen (mg/l)	0-15 (2.71)	0-15 (3.03)
Calcium (mg/l)	3-94.9 (35.35)	1.3-83.9 (27.67)
Magnesium (mg/l)	0.7-58.1 (8.72)	1-49.5 (13.18)
Chloride (mg/l)	0.5-1460 (71.40)	1-3900 (439.36)
Specific Conductance (micromhos/cm)	0.17-5.0 (0.87)	0.15-12.8 (2.65)
Sulfate (mg/l)	0.5-1275 (83.53)	1-2250 (300.11)
Total Alkalinity (mg/l)	46-1465 (247.26)	46-2880 (695.94)
Carbonate Alkalinity (mg/l)	0-776 (34.97)	0-1002 (113.37)
Bicarbonate Alkalinity (mg/l)	46-810 (212.29)	46-2257 (576.77)
Total Hardness (mg/l)	21-441 (134.29)	12-387 (116.77)
Chemical Oxygen Demand (mg/l)	28-466 (123.30)	28-1500 (216.67)
Potassium (mg/l)	2.2-25.8 (10.45)	3.3-5400 (781.36)
Sodium (mg/l)	3.1-102.8 (15.09)	2.8-145.1 (39.66)

variables can be utilized to describe the overall water chemistry optimums for each particular species.

The average mean water temperature for the 4 major species was 22.5°C, while that of the 3 habitat types was 21.7°C. In contrast, the mean water temperature for non-breeding sites was 18.8°C. Larvae were found on only 3 occasions in water below 18°C. Kliewer et al. (1969) tested the relative tolerance of Aedes larvae to various temperatures. Rapid mortality was noted at the extreme ends of the ranges, 0°C and 43°C, respectively. Increases in larval development were noted at temperatures above 18°C, with maximum larval development occurring at a mean of 22.5°C for 1 species, and at 25.7°C for the other species.

Mean pH and DO levels of the major species are reflective of their dominant breeding habitats. The highest recorded mean pH (8.12) and DO (5.06 mg/l) levels were from the artificial container sites, while the lowest mean pH (6.93) and DO (1.27 mg/l) levels were observed from the storm sewer catch basin habitat type. Similarly, total alkalinity, a combination of carbonate and bicarbonate alkalinity, had a positive correlation with pH. Site types with higher alkalinity levels displayed correspondingly higher pH levels and vice versa. No carbonate alkalinity was observed from storm sewer catch basin sites. Petersen and Chapman (1969) observed quite similar relationships between the amounts of bicarbonate and carbonate alkalinity in water samples as compared to the pH levels. On a species basis, they reported the following mean carbonate values: Ae. triseriatus (28.2 mg/l); Cx. restuans (1.20 mg/l) and; Cx. salinarius (0 mg/l). Though their sample size was relatively small, the reported data are consistant with that of this study.

Another direct variable correlation can be seen in the relationship between calcium, magnesium, and total hardness. Ground cavity sites had a high mean level of total hardness (291.33 mg/l) and correspondingly high mean concentrations of calcium (63.62 mg/l) and magnesium (31.52 mg/l), the major constituents of water hardness. In contrast, the total hardness level for the artificial container habitat type was relatively high (219.16 mg/l), while the individual concentrations of calcium (22.77 mg/l) and magnesium (11.06 mg/l) were low. This relationship indicates that other factors contributed to the overall hardness level. In this case, iron compounds added significantly to the overall hardness level. The artificial container sites also had a high mean specific conductance level (4.13 micromhos/cm x 10^3), a value related to the total concentration of ionized substances in the water. This high level is attributed to the high hardness level resulting from the additional iron compounds.

All major species were associated with relatively high chloride levels, with the exception of _Cx. restuans_. The lower mean chloride level (71.40 mg/l) associated with this species can be directly attributed to the corresponding breeding habitats. Both storm sewer and ground cavity habitat types displayed similarly low chloride levels (23.77 mg/l and 46.08 mg/l). A similar situation existed between _Cx. restuans_ and low mean sulfate levels (83.53 mg/l). In several instances, strong correlations existed between chloride and sulfate.

The mean COD value was highest among artificial container sites (301.95 mg/l) and associated species. This high COD value can again be attributed to the oxidizable materials resulting from decomposition of the steel drums. The mean COD of the other 2 habitat types was lower. Again, high levels of potassium (1322.88 mg/l) and sodium (58.02 mg/l)

were associated with the artificial container sites due to decomposition of the steel drums. Because these levels were extremely high, the potassium and sodium levels for the associated species were also high. Low mean potassium (10.45 mg/l) and sodium (15.09 mg/l) levels were associated with Cx. restuans, which predominately bred in storm sewers and ground cavities.

As previously indicated, all non-breeding water collection data were taken from storm sewer catch basin sites. As a rule, the mean variable values for non-breeding collection data were lower than those from the same breeding sites, in which larvae were present. The exceptions were a higher COD level (136.0 mg/l) for non-breeding site data, and relatively equal mean values for pH (6.94) and DO (1.34 mg/l). To further visualize the distribution of variable values, the overall range of each variable was broken down into several sub-ranges. The number of occurrences within each variable sub-range for the major species and habitat types are presented in Tables 6A through 18A. This information is useful in visualizing where the collected data values lie within the overall ranges, and also indicates the number of occurrences at the extreme ends of the range for each variable.

Overall Variable Significance and Rank Order

The multiple linear regression program written for this research was designed to evaluate the impact of each independent variable when removed from the overall equation. By utilizing the R^2 from each regression step output, the significance (F) of each independent variable was calculated using the formula:

$$_{1,\text{residual}} = \frac{(R^2\text{max} - R^2\text{max-1})(\text{residual})}{1 - R^2\text{max}}$$

where the maximum R^2 was contained in the first regression output, which included all independent variables in the equation. From that point on, the independent variables were removed 1 at a time, corresponding to the removal steps in the regression program. Once the significance of each variable was calculated from the R^2 value, it was possible to rank order the variables in terms of significance to the overall equation. The results reflected the relative importance of each variable in relation to the dependent variable, the number of associated mosquito larvae. In some cases, the F values, according to the percentage points of the F distribution, were found significant. In other cases, although not significant, the rank order of variables by F value can still be considered valid in determining the relative effects of each variable on the associated number of larvae.

Separate computer programs were run for each data set. The only variables not included on each surveillance observation were COD, sodium, and potassium, due to the lesser number of observations for these variables. The standard selected variables are rank ordered and the interactions of the independent variables noted.

The first regression program included all site data collected during the research. The variable rank order, by calculated F value and associated statistical output, is presented in Table 17. The rank order of the overall data can be considered indicative of the relative importance of the measured variables in relation to general larval development. In addition to the R^2 used to calculate the F values, other outputs were

Table 17. Variable rank order for overall data.

Variable	F Value	Significance	Correlation Coefficient
pH	2.72	.104	.28
Water Temperature	2.20	.143	.23
Bicarbonate	1.54	.220	.18
Calcium	1.52	.223	-.07
Hardness	1.42	.239	.03
Sulfate	0.63	.431	.19
Magnesium	0.62	.436	.09
Carbonate	0.54	.466	.18
Specific Conductance	0.20	.656	-.07
Chloride	0.12	.728	.15
Dissolved Oxygen	0.02	.885	.22
Overall	1.50	.144	--

obtained from the regression programs. A summary table containing the significance of each variable was outputted, as well as a correlation coefficient matrix, showing the interaction of all variables included in the equation. The correlation coefficients contained in the matrix describe the interrelationships among the independent variables.

When compared with the dependent variable, number of larvae, the highest coefficients correlated to pH (.28) and water temperature (.23). It was also noted from the correlation coefficient matrix that a high degree of correlation existed between total hardness and both calcium (.89) and magnesium (.84). Additionally, a strong correlation was observed between chloride and sulfate (.87). This positive correlation indicates that as the level of 1 variable rises or falls, so does the other. A similar correlation existed between pH and bicarbonate alkalinity (.74). This relationship indicates that bicarbonate alkalinity is quite important in the overall equation because of its effect on pH, the most significant variable in regard to the number of larvae.

Having identified pH, water temperature, and bicarbonate as the variables having the most impact on generalized mosquito breeding, calculations were performed to determine the y-intercept and slope of the corresponding data. A linear regression line was constructed to show the relationship between each variable and the number of mosquito larvae. Regression lines pertaining to the more significant variables for overall and habitat type breeding are contained in Figures 1A through 7A. The correlation coefficients calculated during the formulation of regression lines were done by simple linear regression. The regression lines and associated correlation coefficients directly reflect the effects of a single independent variable on the number of associated larvae.

Regression Analyses of Habitat Types

Another regression program was accomplished for all data collected from the water analyses of storm sewer catch basins. Utilizing the R^2 and the previous formula for F value, the variable rank order presented in Table 18 was obtained. The F values for both water temperature and magnesium were found significant at the .99 confidence level.

From the correlation coefficient matrix, the strongest correlation existed between the number of larvae and water temperature (.39). Positive correlations also existed within the total hardness complex. Calcium (.94), bicarbonate (.90), and magnesium (.82) all displayed high correlations with total hardness. Similarly, bicarbonate correlated with both calcium (.86) and magnesium (.76). As to be expected, a relatively high correlation also existed between calcium and magnesium (.71). These correlations indicate that calcium, magnesium, and bicarbonate alkalinity all contribute to the total hardness of storm sewer catch basin breeding waters. Regression lines for the significant storm sewer catch basin variables are contained in Figures 2A and 3A.

The variable rank order for the artificial container water analyses data are presented in Table 19. The lower overall significance level (.566) resulted in similarly low correlation coefficients. Although the variables had less influence, their importance by rank order can still be considered valid. The 2 most significant variables, bicarbonate (-.17) and calcium (-.10), both displayed negative correlation coefficients, indicating slight limiting effects on mosquito development. The highest positive correlation existed between the number of larvae and magnesium (.18). Calcium and magnesium acted independently on the number of

Table 18. Variable rank order for storm sewer catch basin data.

Variable	F Value	Significance	Correlation Coefficient
Water Temperature	10.38[a]	.002	.39
Magnesium	7.14[a]	.011	-.08
Sulfate	3.03	.090	.05
Bicarbonate	1.67	.205	.16
Hardness	0.88	.355	.08
Calcium	0.45	.508	.06
Chloride	0.25	.621	-.09
pH	0.12	.736	.05
Specific Conductance	0.12	.736	-.09
Dissolved Oxygen	0.01	.910	-.06
Carbonate	0.00	1.000	.00
Overall	2.25	.270	--

[a]Significant at the .99 confidence level

Table 19. Variable rank order for artificial container data.

Variable	F Value	Significance	Correlation Coefficient
Bicarbonate	2.25	.160	-.17
Calcium	1.79	.208	-.10
pH	1.56	.239	.09
Magnesium	0.76	.407	.18
Sulfate	0.66	.437	.01
Hardness	0.47	.511	.09
Dissolved Oxygen	0.33	.583	.13
Water Temperature	0.20	.666	-.13
Chloride	0.06	.806	-.01
Carbonate	0.03	.879	-.10
Specific Conductance	0.00	.966	-.04
Overall	0.89	.566	--

larvae, as little correlation existed between the 2. Although specific conductance ranked last in order of importance, several variables correlated with it. Sulfate (.88), chloride (.78), magnesium (.73), and bicarbonate (.71), all showed relatively high correlation coefficients with specific conductance. All of these variables contribute to the total dissolved particles as measured by specific conductance. High correlations also existed between sulfate and chloride (.83) and magnesium (.81). Although chloride and sulfate had low correlations with the number of larvae, they had a combined effect on magnesium, which in turn had the highest positive correlation with the number of larvae. Regression lines showing the effects of these variables are presented in Figures 1A through 7A.

The regression program for the ground cavity water collection data showed all summary table significance levels equal to 1.000, indicating the sample size was too small for statistical manipulation. Individual variable ranges and means (Table 16) can still be considered valid for habitat type comparative purposes.

Regression Analyses of Individual Species

Multiple linear regression programs were run for the 4 major species of concern. The data sets for the 7 incidental species were not considered valid, due to insufficient amounts of data. The variable rank order presented in Table 20 was observed from the regression program run for the *Ae. triseriatus* environmental and water chemistry data. The F value for specific conductance proved significant at the .95 confidence level. It should again be noted that *Ae. triseriatus* was collected only from the artificial container habitat type.

Table 20. Variable rank order for *Aedes triseriatus* data.

Variable	F Value	Significance	Correlation Coefficient
Specific Conductance	5.91[a]	.057	-.48
Dissolved Oxygen	3.27	.135	.19
Calcium	2.29	.199	.03
Hardness	2.28	.201	.01
Magnesium	2.22	.206	-.05
pH	1.26	.329	.23
Carbonate	1.15	.350	-.13
Chloride	0.45	.550	-.28
Water Temperature	0.13	.742	-.07
Sulfate	0.09	.789	-.31
Bicarbonate	0.09	.789	-.42
Overall	1.53	.293	--

[a]Significant at the .95 confidence level

The highest correlation within the Ae. triseriatus matrix existed between the number of larvae and specific conductance (-.48). This negative correlation indicates that specific conductance has a limiting effect on larval development. Correlation interrelationships existed between sulfate and specific conductance (.74), and sulfate and hardness (.92). These relationships indicate that sulfate is an important contributing factor to the overall effect of specific conductance and that hardness also influences this effect. Ae. triseriatus was the only species to display a positive correlation to DO (.19), indicating that as DO values increase, so do the number of associated larvae. Similarly, the artificial container habitat type had the highest mean DO value (5.06 mg/l) of all types sampled. Regression lines for the more important variables influencing Ae. triseriatus development are presented in Figures 8A and 9A.

The regression program for the Cx. pipiens data resulted in the variable rank order contained in Table 21. Again, the low significance of the overall equation (.949) resulted in correspondingly low correlation coefficients. Additional sampling would serve to intensify the variable correlations and their interrelationships.

From the correlation coefficient matrix, bicarbonate (-.14) displayed the highest correlation when compared to the dependent variable, with chloride (-.12) ranking second, though somewhat less significant. Both of these variables acted to limit Cx. pipiens development as their values increased. Several variable interactions were noted. Bicarbonate displayed strong correlations with pH (.87), and specific conductance (.85). Similarly, specific conductance, which ranked fourth in order of

Table 21. Variable rank order for *Culex pipiens* data.

Variable	F Value	Significance	Correlation Coefficient
Bicarbonate	1.88	.193	-.14
Magnesium	1.13	.308	.04
Sulfate	0.78	.397	-.09
Specific Conductance	0.73	.411	-.06
Chloride	0.43	.526	-.12
Hardness	0.30	.597	.03
Dissolved Oxygen	0.30	.597	-.04
Calcium	0.26	.625	-.01
pH	0.11	.750	-.08
Carbonate	0.05	.836	-.07
Water Temperature	0.01	.921	-.07
Overall	0.38	.949	--

importance, correlated highly with sulfate (.93), chloride (.83), pH (.80), and magnesium (.80). Sulfate also showed high correlations to chloride (.85) and magnesium (.84), while magnesium and chloride together displayed a relatively high correlation (.72).

Bicarbonate alkalinity, the variable having the most significant effect on Cx. pipiens development, also influences pH and specific conductance, showing strong positive correlations. The remaining top 5 variables, magnesium, sulfate, specific conductance, and chloride, all displayed a degree of interaction with one another.

The variable rank order for the Cx. restuans data is presented in Table 22. The calculated F value for chloride was found significant at the .99 confidence level, while the F value for sulfate was significant at the .95 confidence level. Chloride displayed a slight negative correlation coefficient (-.08), indicating a limiting effect on Cx. restuans development. Chloride correlated highly with sulfate (.95) and specific conductance (.88), while sulfate and specific conductance also correlated highly (.96). The remaining top 6 variables were from the total hardness complex and a number of expected interactions were noted. Specific conductance ranked last in order of importance, but its interactions with other variables, namely chloride and sulfate, influenced the development of Cx. restuans. Regression lines for the variables having the most effect on Cx. restuans development are contained in Figures 10A and 11A.

The variable rank order for the Cx. salinarius water chemistry data is contained in Table 23. In the case of Cx. salinarius, additional sampling would tend to enhance the variable correlation coefficients and their resulting interactions with one another. The majority of correlation coefficients were negative and therefore tended to limit larval

Table 22. Variable rank order for *Culex restuans* data.

Variable	F Value	Significance	Correlation Coefficient
Chloride	8.14[a]	.012	-.08
Sulfate	7.41[b]	.016	.14
Carbonate	3.70	.076	.58
Bicarbonate	2.21	.165	.53
Calcium	1.51	.246	.54
Hardness	1.09	.322	.48
Dissolved Oxygen	0.39	.550	-.16
Magnesium	0.36	.565	.36
Water Temperature	0.17	.694	-.30
pH	0.01	.941	.27
Specific Conductance	0.00	.988	.26
Overall	4.76	.001	--

[a]Significant at the .99 confidence level

[b]Significant at the .95 confidence level

Table 23. Variable rank order for *Culex salinarius* data.

Variable	F Value	Significance	Correlation Coefficient
Calcium	1.95	.185	-.05
Hardness	1.58	.230	-.19
Dissolved Oxygen	1.27	.282	-.22
Specific Conductance	0.73	.412	-.03
Magnesium	0.42	.530	-.19
Carbonate	0.27	.618	-.14
Water Temperature	0.14	.717	-.10
Bicarbonate	0.09	.771	-.13
pH	0.00	.958	-.23
Chloride	0.00	.961	.02
Sulfate	0.00	.963	-.02
Overall	0.68	.739	--

development. These results can be interpreted to indicate that _Cx. salinarius_ is more sensitive to the chemical nature of its breeding waters than the other species studied. Calcium and hardness had the most influence on larval development. In turn, a relatively high correlation existed between these 2 variables (.76). A similarly high correlation existed between magnesium and hardness (.82).

CONCLUSIONS

Of the 11 mosquito species collected, 4 were identified as proven or suspected vectors. St. Louis and CE are the primary disease threats associated with these mosquito species. Larval and adult mosquito control efforts should be maximized during the critical breeding months of July, August, and September. The incidental species, Ps. columbiae and Ps. howardii, are determined to be new Tippecanoe County collection records. Single specimens of each species were taken.

Cx. restuans was the dominant early season species. Although not considered a primary vector of SLE, the role of Cx. restuans is not clearly defined. Early season dominance may serve to build and sustain SLE viral activity in the avian reservoir population, until primary vector species become active. Cx. restuans may also be less incriminated as a primary vector, due to low population levels in August and September. It is suspected that this species plays an important role in the overall SLE transmission cycle. Early season reductions in Cx. restuans populations could decrease associated viral activity, thereby effectively reducing later season disease occurrences.

The 2 most significant variables associated with Cx. restuans development were chloride and sulfate. The mean chloride value was 71.40 mg/l, by far the lowest of the 4 major species. The associated correlation coefficient of -.08, indicates that the presence of high chloride

levels tend to limit the breeding of Cx. restuans. The highest observed chloride level associated with this species was 1460 mg/l, as compared to 3900 mg/l for the other major species. These results indicate that the addition of chloride compounds to breeding waters to produce levels above 1500 mg/l, could effectively limit Cx. restuans population development. Sulfate had a mean value of 83.53 mg/l, again, the lowest of all major species encountered. However, the positive correlation coefficient of .14 indicates that higher levels of sulfate would result in higher population levels. Therefore, a reduction in existing sulfate levels would further limit the development of the species. Chloride and sulfate had a correlation coefficient of .95, indicating a significant combined effect on Cx. restuans development.

The majority of Cx. salinarius variable correlation coefficients were negative, and indicated a trend toward limiting larval development. The 2 most influential variables, calcium and hardness, were both limiting, and together had a correlation coefficient of .76. Similarly, magnesium and hardness also correlated highly. These results indicate that the entire total hardness complex may have a limiting effect on the development of this species. It is suspected that hardness levels approaching 400 mg/l would severly limit Cx. salinarius development.

An additional vector-borne disease threat was manifested by the development of large Ae. triseriatus populations from artificial container habitats. Specific conductance was the most influential variable, with a correlation coefficient of -.48, indicating a limiting effect on larval development. The mean specific conductance value was the highest of all monitored species. Ae. triseriatus can tolerate relatively higher

mean levels of dissolved salts, but at some level in excess of 7.50 micromhos/cm x 10^3, these salts become limiting.

Ae. triseriatus was the only major species to show a positive correlation with DO. This species was never found breeding in water with less than a 3 mg/l DO level. This fact indicates that the elimination of DO from potential breeding sites would severely limit development of the species.

Bicarbonate alkalinity was the most influential variable with regard to Cx. pipiens development. It had a mean value of 527.97 mg/l. The data suggests that bicarbonate values approaching 2500 mg/l become severely limiting. Bicarbonate also displayed strong correlations with pH and specific conductance. These interactions indicate that increased levels of total dissolved salts may also have a combined limiting effect on the development of Cx. pipiens.

The storm sewer catch basin habitat type was concluded to be the most important monitored. The persistence of Cx. pipiens in storm sewer catch basins is of great concern. Together with its proven vector potential and observed domesticity, Cx. pipiens provides the single most significant disease threat to the populace of this area.

Of the 9 storm sewer catch basins monitored, only site 18 did not produce mosquito larvae. All factors considered, this catch basin was virtually identical to the others except for physical construction. A constant water level of 25 cm was maintained at a distance of 1.5 m below pavement level, considerably lower than the 1 m of the next deepest catch basin. Since no unusual circumstances were observed, and since other catch basins in this part of the city produced larvae, it is

concluded that the increased depth of this catch basin acted to markedly decrease the oviposition appeal of gravid female mosquitoes. This fact should be considered during the planning and construction of new storm sewer systems.

The variables which had the most influence on larval development in storm sewer catch basin habitats were water temperature and magnesium. Water temperature displayed a positive correlation coefficient, indicating that an increase in breeding site water temperature results in increased larval development. The mean water temperature for storm sewer catch basin breeding collections was 21.3°C, while that of non-breeding collections was 18.8°C. Larvae were found in this habitat type on only 3 occasions in water below 18°C, indicating that a 3-4°C rise in water temperature has a significant positive effect on larval development. A water temperature of near 18°C seemed to trigger larval activity in this specific habitat type.

The mean magnesium level for storm sewer catch basins was the lowest of all habitat types. Several strong correlations existed with magnesium and the total hardness complex in general. These interrelationships indicate that the total hardness complex, as a whole, has an important limiting effect on mosquito development in the storm sewer catch basin habitat type. The data suggests that even relatively small increases in total hardness complex components would further limit species development.

Bicarbonate alkalinity had the most influence on the artificial container habitat type. The negative correlation coefficient reflects a limiting effect on larval development. The mean bicarbonate alkalinity

value was the highest of all habitats sampled. In turn, Ae. triseriatus, the dominant species in this habitat, demonstrated an ability to tolerate higher alkalinity levels than other monitored species. These levels, however, become limiting at the upper end of the range. The data suggests that alkalinity levels in excess of 3000 mg/l would severely limit continued Ae. triseriatus development.

The ground cavity sites displayed the greatest species diversity, producing 9 of the 11 species collected. The early season dominance of Cx. restuans from this habitat type must be taken into account when considering its potential role in disease transmission. Linear regression analysis of the ground cavity variables was not feasible due to the limited sample size. The individual variable ranges and means reported earlier are considered valid for habitat type comparisons. The ground cavity sites had the highest mean values for calcium, magnesium, and total hardness of the 3 site types monitored. The conspicuous absence of Cx. salinarius is directly attributed to the limiting effects of these high value hardness components.

The variable ranges and means reported in this study are unique to the specific breeding situations monitored. Ranges and means may vary for individual species when sampled from varying geographic areas and habitats. It is concluded from this work that future researchers would be well advised to concentrate on a single species for studies of this type, or at most, a single habitat type.

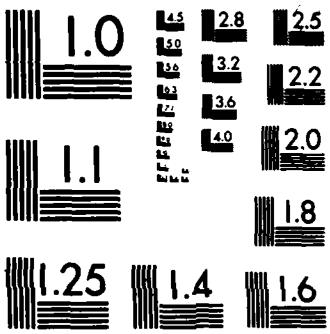
The major species monitored during this study displayed relatively high tolerances to a wide range of variable values. The information collected here provides a foundation for comparative studies conducted in a

similar manner. Further studies along these lines of investigation may eventually lead to a more complete description of larval habitats and associated species. Complete understanding of this aspect of mosquito bionomics will lead to more efficient and lasting control measures.

AD-A128 506 MOSQUITO PRODUCTION AND ASSOCIATED ENVIRONMENTAL AND CHEMICAL PARAMETERS OF BREEDING WATERS(U) AIR FORCE INST OF TECH WRIGHT-PATTERSON AFB OH T J KASA MAY 83 2/2

UNCLASSIFIED AFIT/CI/NR-83-13T F/G 6/3 NL

END
FILMED
DTIC



MICROCOPY RESOLUTION TEST CHART
NATIONAL BUREAU OF STANDARDS-1963-A

LIST OF REFERENCES

LIST OF REFERENCES

American Public Health Association. 1981. Standard methods for the examination of water and wastewater. American Public Health Association. Washington, D.C. 1034 pp.

Barton, W. I. 1977. A special study to show larval densities in relation to water depth. Mosq. News. 37(2): 130.

Bast, T. F. 1963. Chemical nature of mosquito breeding waters. Proc. 50th Ann. Meet. N. J. Mosq. Exterm. Assn. 335-339.

Bast, T. F. 1964. Further studies on the chemical nature of mosquito breeding waters in New Jersey. Proc. 51st Ann. Meet. N. J. Mosq. Exterm. Assn. 129-133.

Bast, T. F. 1965. Sulfate, phosphate, and chloride as limiting factors in mosquito production. Proc. 52nd Ann. Meet. N. J. Mosq. Exterm. Assn. 193-199.

Brooks, I. C. 1947. The mosquitoes of Tippecanoe County: Their habitats and environments. Master of Science Thesis. Purdue University. 42 pp.

Carpenter, S. J. and W. J. LaCasse. 1955. Mosquitoes of North America north of Mexico. Univ. of Calif. Press, Berkely and Los Angeles. 360 pp.

Chamberlain, R. W., R. H. Gogel, and W. D. Sudia. 1966. Experimental vector studies with strains of St. Louis encephalitis virus isolated from mosquitoes during the 1964 epidemics. J. Med. Entomol. 3(3-4): 268.

Chubachi, R. 1976. The efficiency of the dipper in sampling of mosquito larvae and pupae under different conditions. Science Reports of the Tohoku University, IV. 37(2): 145-149.

Earle, H. H. 1956. Automatic device for the collection of aquatic specimens. J. Econ. Entomol. 49(2): 261-262.

Hagstrum, D. W. and S. E. Gunstream. 1971. Salinity, pH, and organic nitrogen of water in relation to presence of mosquito larvae. Ann. Entomol. Soc. Amer. 64(2): 465-467.

Hart, J. W. 1942. Preliminary studies of the mosquitoes of Indiana. Bachelor of Science Thesis. Purdue University. 47 pp.

Hayes, J. 1973. A study of the Culex pipiens complex in the Ohio-Mississippi River Basin in relation to St. Louis encephalitis. Mosq. News. 33(3): 447-454.

Karkatzke, J. T. 1980. Effect of sodium chloride on larval snow-melt Aedes (Diptera:Culicidae). Mosq. News 40(2): 153-160.

Kardatzke, J T. 1981. Effect of addition of calcium chloride to larval medium on survival of immature snow-melt Aedes (Diptera:Culicidae). Mosq. News. 41(1): 11-12.

King, W. V., G. H. Bradley, C. N. Smith, and W. C. McDuffie. 1960. A handbook of the mosquitoes of the southeastern United States. Agr. Handbook No. 173, Agf. Res. Serv., U.S.D.A., Washington, D.C., 118 pp.

Kliewer, J. W., M. Takeshi, and K. E. White. 1964. Temperature and salinity relationships of Aedes nigromaculis and Ae. melanimon. Proc. Calif. Mosq. Cont. Assn. 32: 42-45.

Knight, K. L. and C. Henderson. 1967. Flight periodicity of Aedes vexans (Meigen) (Diptera:Culicidae). Georgia Entomol. Soc. J. 2(3): 63-68.

Kokernot, R. H., J. Hayes, N. J. Rose, and T. H. Work. 1967. St. Louis encephalitis in McLeansboro, Illinois, 1964. J. Med. Entomol. 4(3): 255-260.

Lang, J. T., D. D. Pinkovsky, and R. J. McKenna. 1981. Mosquito vectors collected at CONUS USAF installations and mosquito-borne disease data. USAF School of Aerospace Medicine Technical Report 81-36, Brooks AFB, Texas. 222 pp.

Lindsey, A. A., D. V. Schmelz, and S. A. Nichols. 1969. Natural areas in Indiana and their preservation. Indiana Natural Areas Survey, Purdue University, Lafayette, Indiana. 594 pp.

Loomis, E. C. 1959. The function of larval surveys in the California encephalitis surveillance program. Proc. Calif. Mosq. Cont. Assn. 21: 64-67.

Loor, K. A. and G. R. DeFoliart. 1970. Field observations on the biology of Aedes triseriatus. Mosq. News. 30(1): 60-64.

Lumsden, L. L. 1959. St. Louis encephalitis in 1933: Observations on epidemiological features. Public Health Report. 73: 340-353.

Mack, T. M., B. F. Brown, W. D. Sudia, J. C. Todd,H. Maxfield, and P. H. Coleman. 1967. Investigation of an epidemic of St. Louis encephalitis in Danville, Kentucky, 1964. J. Med. Entomol. 4(1): 70-76.

Maddock, D. R., M. Elmore, Jr., and J. F. Schoof. 1963. Preliminary tests with DDVP vapor for the control of Culex pipiens quinquefasciatus in catch basins. Mosq. News. 23(1): 69-75.

Marshall, A. L. 1965. In: Encephalitis Surveillance Summary, Morbidity and Mortality Weekly Reports. 13(41): 366.

McIver, S. B. 1969. Notes on the biology of Culex territans Walker. Mosq. News. 29(1): 135-136.

Means, R. G. 1965. Culex territans Walker biting man in nature. Mosq. News. 25(4): 489.

Newhouse, V. F. and R. E. Siverly. 1966a. The Culex pipiens complex in southern Indiana. Mosq. News. 25(4): 489-490.

Newhouse, V. F. and R. E. Siverly. 1966b. St. Louis encephalitis virus from mosquitoes in southwestern Indiana, 1964. J. Med. Entomol. 3(3-4): 340-341.

Peterson, J. J. and D. M. Rees. 1966. Selective oviposition response of Aedes dorsalis and Aedes nigromaculis to soil salinity. Mosq. News. 26(2): 169-174.

Peterson, J. J. and H. C. Chapman. 1969. Chemical factors of water in tree holes and related breeding of mosquitoes. Mosq. News. 29(1): 29-36.

Peterson, J. J. and O. R. Willis. 1971. Effects of salinity on site selection by ovipositing tree hole mosquitoes in Louisiana. Mosq. News. 31(3): 352-355.

Razenhofer, E. R., E. R. Alexander, L. D. Beadle, A. Bernstein, and R. C. Pickard. 1957. St. Louis encephalitis in Calvert City, Kentucky, 1955. An epidemiologic study. Amer. J. Hyg. 65(2): 147-161.

Service, M. W. 1976. Mosquito ecology. Field sampling methods. Appl. Sci. Publ., London. 583 pp.

Shriver, D. and W. E. Bickley. 1964. The effect of temperature on hatching of eggs of the mosquito, Culex pipiens quinquefasciatus Say. Mosq. News. 24(1): 137-140.

Sinsko, M. J. and G. B. Craig. 1979. Dynamics of an isolated population of Aedes triseriatus (Diptera:Culicidae) I. Population size. J. Med. Entomol. 15(1): 89-98.

Siverly, R. E. 1972. Mosquitoes of Indiana. Indiana State Board of Health. Indianapolis. 126 pp.

Smith, L. W. 1969. The relationship of mosquitoes to oxidation lagoons in Columbia, Missouri. Mosq. News. 29(4): 556-563.

Sudia, W. D., E. Fowinkle, and P. H. Coleman. 1967. St. Louis encephalitis in Memphis, Tennessee, 1964. J. Med. Entomol. 4(1): 77-79.

Watts, D. M., S. Pantuwatana, G. R. DeFoliart, T. M. Yuill, and W. H. Thompson. 1973. Transovarial transmission of LaCrosse virus (California encephalitis group) in the mosquito Aedes triseriatus. Science. 182(7): 1440-1441.

APPENDIX

Table 1A. Aedes triseriatus surveillance data summary.

Month	No. Collections	No. Specimens	Average No. Specimens per Collection
May	2	45	22.50
Jun	0	0	0
Jul	11	512	46.55
Aug	9	413	45.89
Sep	8	149	18.63
Oct	0	0	0
Total	30	1119	37.30

Table 2A. Culex pipiens surveillance data summary.

Month	No. Collections	No. Specimens	Average No. Specimens per Collection
May	0	0	0
Jun	2	50	25.00
Jul	10	149	14.90
Aug	23	373	16.22
Sep	19	332	17.47
Oct	2	11	5.50
Total	56	915	16.34

Table 3A. *Culex restuans* surveillance data summary.

Month	No. Collections	No. Specimens	Average No. Specimens per Collection
May	5	270	54.00
Jun	12	445	37.08
Jul	14	415	29.64
Aug	15	238	15.87
Sep	5	22	4.40
Oct	0	0	0
Total	51	1390	27.26

Table 4A. *Culex salinarius* surveillance data summary.

Month	No. Collections	No. Specimens	Average No. Specimens per Collection
May	0	0	0
Jun	4	52	13.00
Jul	10	181	18.10
Aug	17	372	21.88
Sep	21	529	25.19
Oct	4	99	24.75
Total	56	1233	22.02

Table 5A. Summary of major species association showing the % of times a species was collected by itself or with other species.

Species	%			
	Aedes triseriatus	Culex pipiens	Culex restuans	Culex salinarius
Aedes triseriatus	57	20	6	14
Culex pipiens	37	9	47	68
Culex restuans	10	43	39	38
Culex salinarius	27	68	41	14

Table 6A. Summary showing the number of water samples having specific temperatures (°C) and containing larvae of the 4 major study species.

Species	No. Occurrences per Range					
	13-15	16-18	19-21	22-24	25-27	28-30
<u>Aedes</u> <u>triseriatus</u>	2	6	2	11	9	0
<u>Culex</u> <u>pipiens</u>	0	6	13	22	10	5
<u>Culex</u> <u>restuans</u>	2	8	16	16	8	1
<u>Culex</u> <u>salinarius</u>	0	10	13	19	10	4
Total	4	30	44	68	37	10
% of Total	2.1	15.5	22.8	35.2	19.2	5.2

Table 7A. Summary showing the number of water samples having specific hydrogen ion concentrations (pH units) and containing larvae of the 4 major study species.

Species	No. Occurrences per Range					
	6.0-6.5	6.6-7.0	7.1-7.5	7.6-8.0	8.1-8.5	8.6-9.0
Aedes triseriatus	0	0	0	3	25	2
Culex pipiens	2	22	11	5	14	2
Culex restuans	3	21	19	5	3	0
Culex salinarius	5	21	8	3	14	5
Total	10	64	38	16	56	9
% of Total	5.2	33.1	19.7	8.3	29.0	4.7

Table 8A. Summary showing the number of water samples having specific concentrations of dissolved oxygen (mg/l) and containing larvae of the 4 major study species.

Species	No. Occurrences per Range					
	0-1	2-3	4-5	6-7	8-9	10+
Aedes triseriatus	0	1	21	5	1	2
Culex pipiens	21	16	14	3	0	2
Culex restuans	21	18	9	0	0	3
Culex salinarius	22	14	14	2	0	4
Total	64	49	58	10	1	11
% of Total	33.1	25.4	30.1	5.2	0.5	5.7

Table 9A. Summary showing the number of water samples having specific concentrations of calcium (mg/l) and containing larvae of the 4 major study species.

Species	No. Occurrences per Range						
	0-10	11-20	21-30	31-40	41-50	51-75	76+
Aedes triseriatus	10	13	2	3	0	1	1
Culex pipiens	7	12	20	7	2	3	0
Culex restuans	6	7	11	6	9	4	4
Culex salinarius	11	9	17	6	5	3	2
Total	34	41	50	22	16	11	7
% of Total	18.8	22.6	27.6	12.2	8.8	6.1	3.9

Table 10A. Summary showing the number of water samples having specific concentrations of magnesium (mg/l) and containing larvae of the 4 major study species.

Species	No. Occurrences per Range						
	0-10	11-20	21-30	31-40	41-50	51-75	76+
Aedes triseriatus	16	11	3	0	0	0	0
Culex pipiens	36	7	6	0	2	0	0
Culex restuans	37	3	5	1	0	1	0
Culex salinarius	36	6	6	0	5	0	0
Total	125	27	20	1	7	1	0
% of Total	69.1	14.9	11.0	0.6	3.8	0.6	0

Table 11A. Summary showing the number of water samples having specific concentrations of chloride (mg/l) and containing larvae of the 4 major study species.

Species	No. Occurrences per Range						
	1-5	6-10	11-25	26-50	51-250	251-500	501+
<u>Aedes</u> <u>triseriatus</u>	0	0	3	5	8	8	6
<u>Culex</u> <u>pipiens</u>	10	10	16	3	1	5	11
<u>Culex</u> <u>restuans</u>	11	11	14	6	6	0	3
<u>Culex</u> <u>salinarius</u>	11	8	11	3	8	5	10
Total	32	29	44	17	23	18	30
% of Total	16.6	15.0	22.9	8.8	11.9	9.3	15.5

Table 12A. Summary showing the number of water samples having specific values for specific conductance (micromhos/cm x 10^3) and containing larvae of the 4 major study species.

Species	No. Occurrences per Range					
	0.1-0.2	0.3-0.5	0.6-1.0	1.1-3.0	3.1-5.0	5.1+
Aedes triseriatus	1	1	1	10	10	7
Culex pipiens	22	18	2	1	0	13
Culex restuans	19	16	7	4	2	3
Culex salinarius	21	11	3	3	8	10
Total	63	46	13	18	20	33
% of Total	32.6	23.8	6.7	9.3	10.4	17.2

Table 13A. Summary showing the number of water samples having specific concentrations of sulfate (mg/l) and containing larvae of the 4 major study species.

Species	No. Occurrences per Range						
	1-10	11-25	26-50	51-100	101-500	501-1000	1000+
Aedes triseriatus	0	0	8	8	8	1	5
Culex pipiens	14	21	2	3	3	3	10
Culex restuans	14	20	7	5	1	1	3
Culex salinarius	11	18	6	2	8	3	8
Total	39	59	23	18	20	8	26
% of Total	20.2	30.6	11.9	9.3	10.4	4.1	13.5

Table 14A. Summary showing the number of water samples having specific total alkalinity concentrations (mg/l) and containing larvae of the 4 major study species.

Species	No. Occurrences per Range					
	1-50	51-100	101-200	201-500	501-1000	1000+
Aedes triseriatus	0	0	2	5	6	17
Culex pipiens	2	19	11	6	2	16
Culex restuans	2	16	14	13	1	5
Culex salinarius	3	18	6	8	1	20
Total	7	53	33	32	10	58
% of Total	3.6	27.5	17.1	16.5	5.2	30.1

Table 15A. Summary showing the number of water samples having specific carbonate alkalinity concentrations (mg/l) and containing larvae of the 4 major study species.

Species	No. Occurrences per Range						
	0	1-50	51-100	101-200	201-500	501-1000	1000+
Aedes triseriatus	10	0	6	3	3	6	2
Culex pipiens	42	0	0	5	5	2	2
Culex restuans	45	0	0	3	2	1	0
Culex salinarius	42	0	0	6	3	3	2
Total	139	0	6	17	13	12	6
% of Total	72.0	0	3.1	8.8	6.7	6.3	3.1

Table 16A. Summary showing the number of water samples having specific bicarbonate alkalinity concentrations (mg/l) and containing larvae of the 4 major study species.

	No. Occurrences per Range					
	1-50	51-100	101-200	201-500	501-1000	1000+
Aedes triseriatus	0	0	2	11	3	14
Culex pipiens	2	19	11	6	2	16
Culex restuans	2	16	14	13	5	1
Culex salinarius	3	18	6	8	5	16
Total	7	53	33	38	15	47
% of Total	3.6	27.5	17.1	19.7	7.8	24.3

Table 17A. Summary showing the number of water samples having specific total hardness concentrations (mg/l) and containing larvae of the 4 major study species.

Species	No. Occurrences per Range						
	1-25	26-50	51-75	76-100	101-125	126-150	150+
Aedes _triseriatus_	3	9	3	2	5	2	6
Culex _pipiens_	3	2	8	19	10	5	9
Culex _restuans_	3	3	6	13	5	6	15
Culex _salinarius_	3	3	8	18	10	3	11
Total	12	17	25	52	30	16	41
% of Total	6.2	8.8	13.0	26.9	15.5	8.3	21.3

Table 18A. Summary showing the number of water samples having specific chemical oxygen demand levels (mg/l) and containing larvae of the 4 major study species.

Species	No. Occurrences per Range					
	0-50	51-100	101-200	201-300	301-400	400+
Aedes triseriatus	0	0	3	11	2	1
Culex pipiens	15	8	6	6	2	5
Culex restuans	16	7	5	3	2	1
Culex salinarius	11	10	5	10	3	5
Total	42	25	19	30	9	12
% of Total	30.7	18.2	13.9	21.8	6.6	8.8

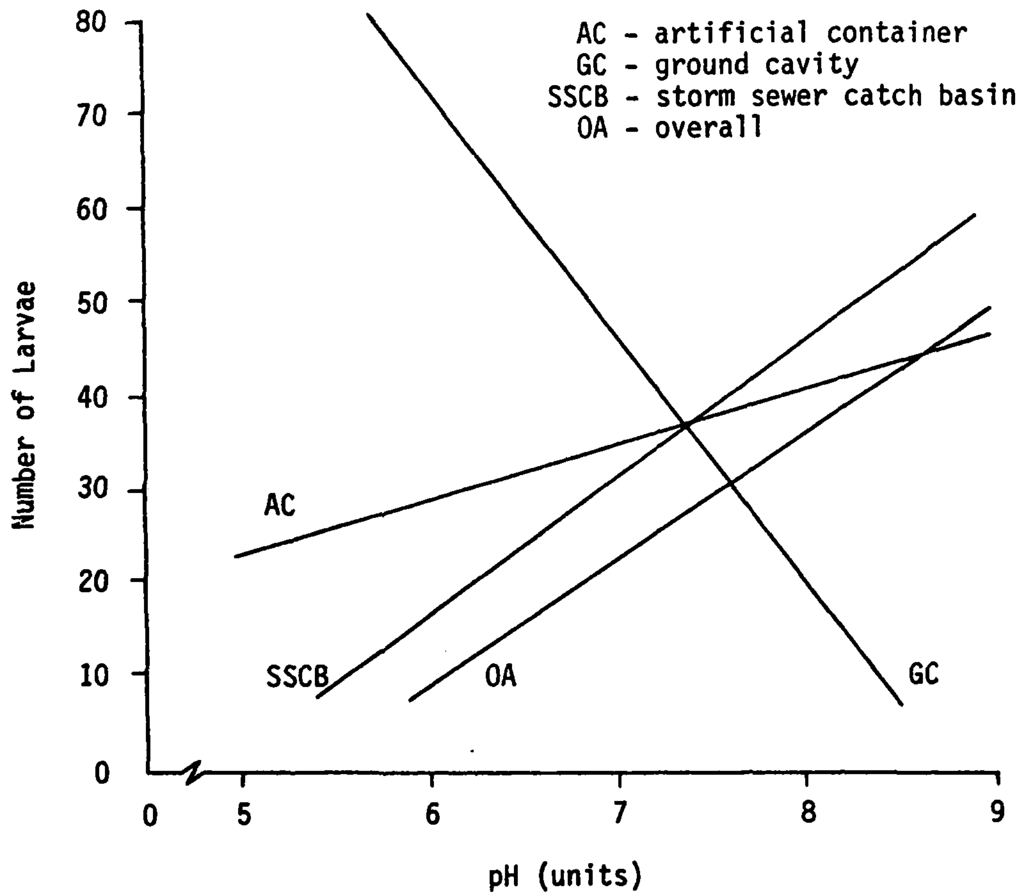


Figure 1A. Regression line comparison of the effects of pH on overall and habitat type larval development.

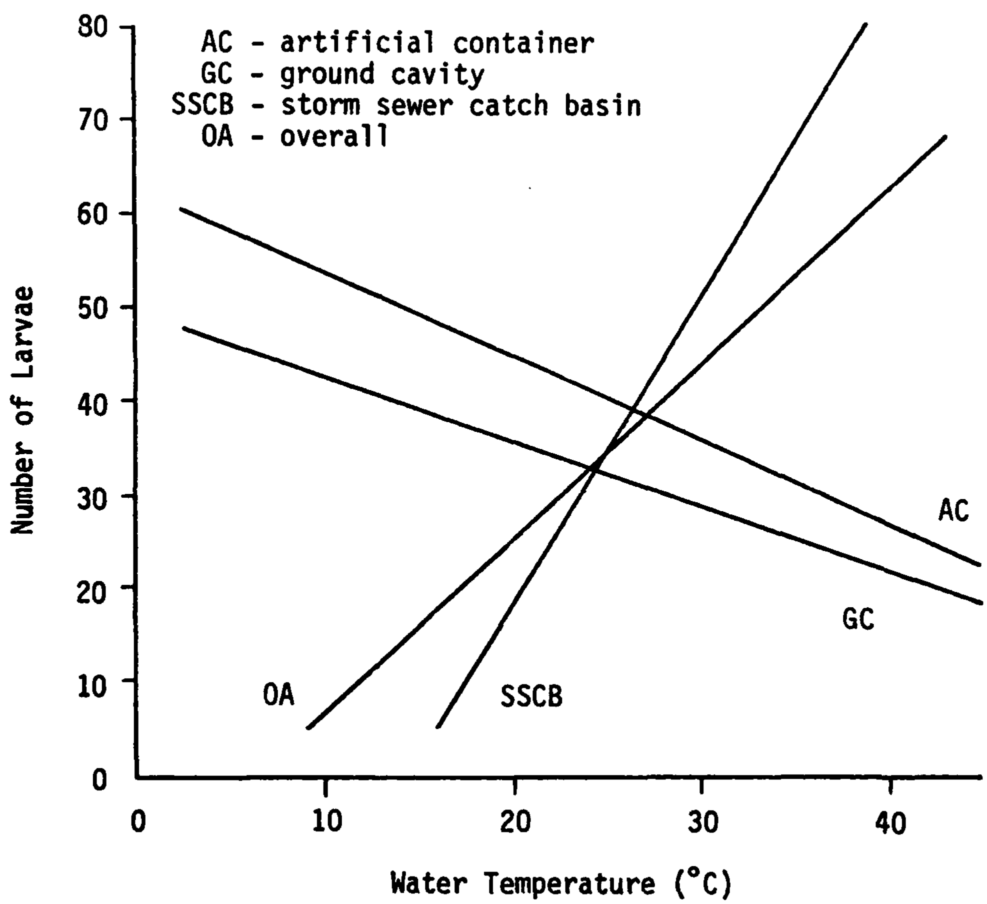


Figure 2A. Regression line comparison of the effects of water temperature on overall and habitat type larval development.

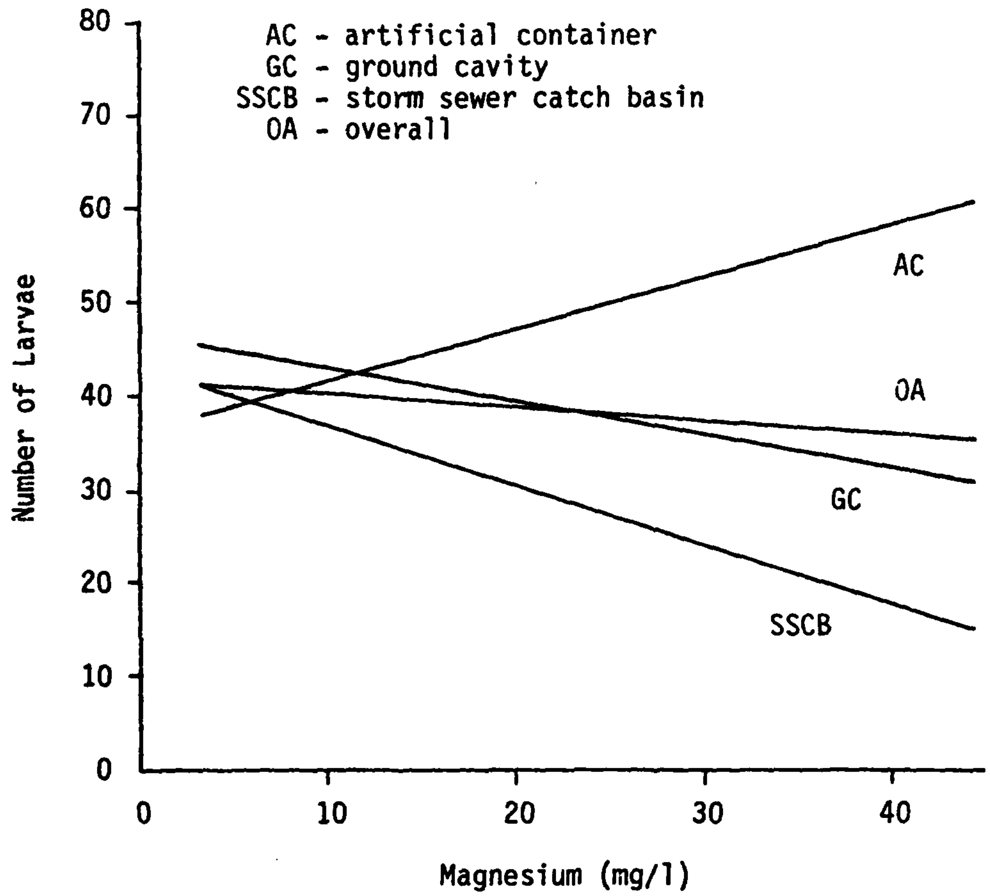


Figure 3A. Regression line comparison of the effects of magnesium on overall and habitat type larval development.

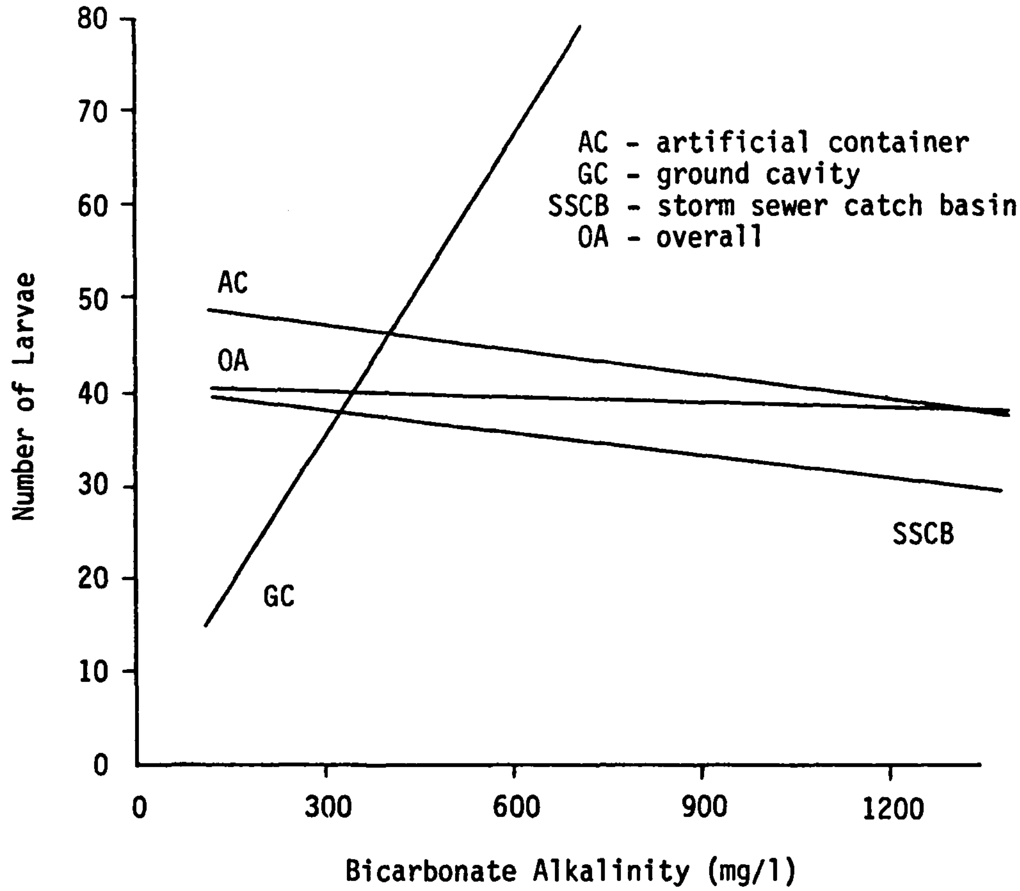


Figure 4A. Regression line comparison of the effects of bicarbonate alkalinity on overall and habitat type larval development.

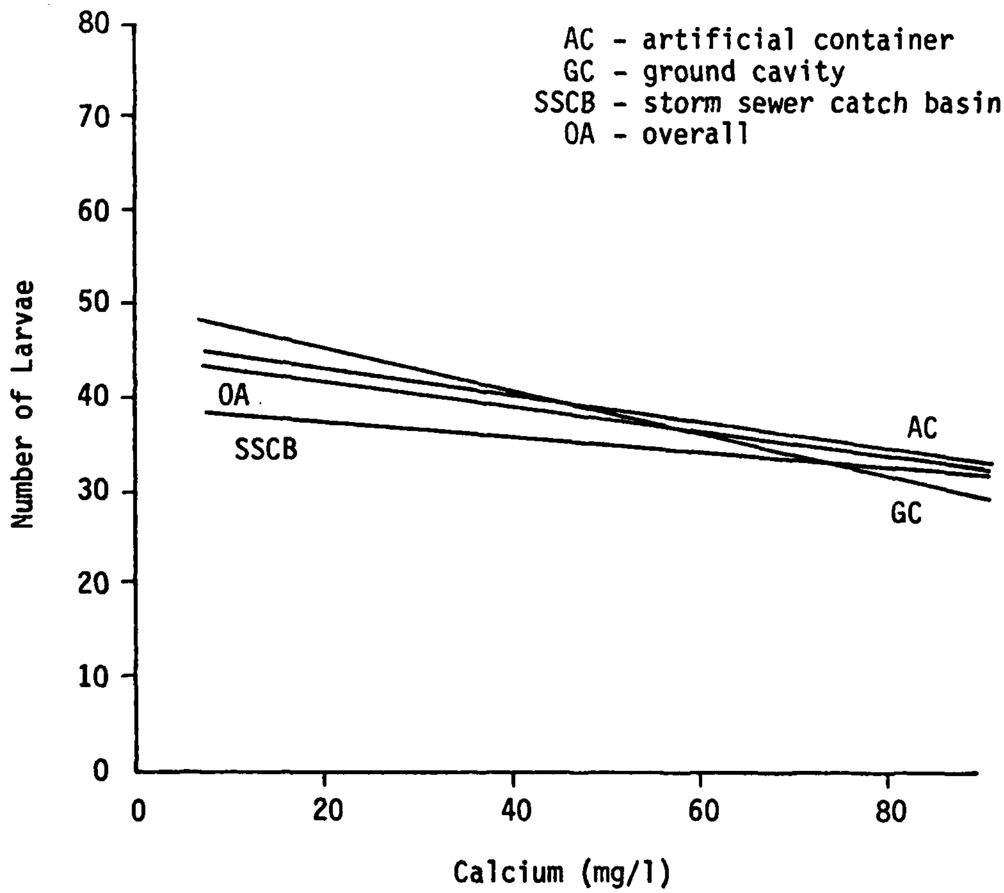


Figure 5A. Regression line comparison of the effects of calcium on overall and habitat type larval development.

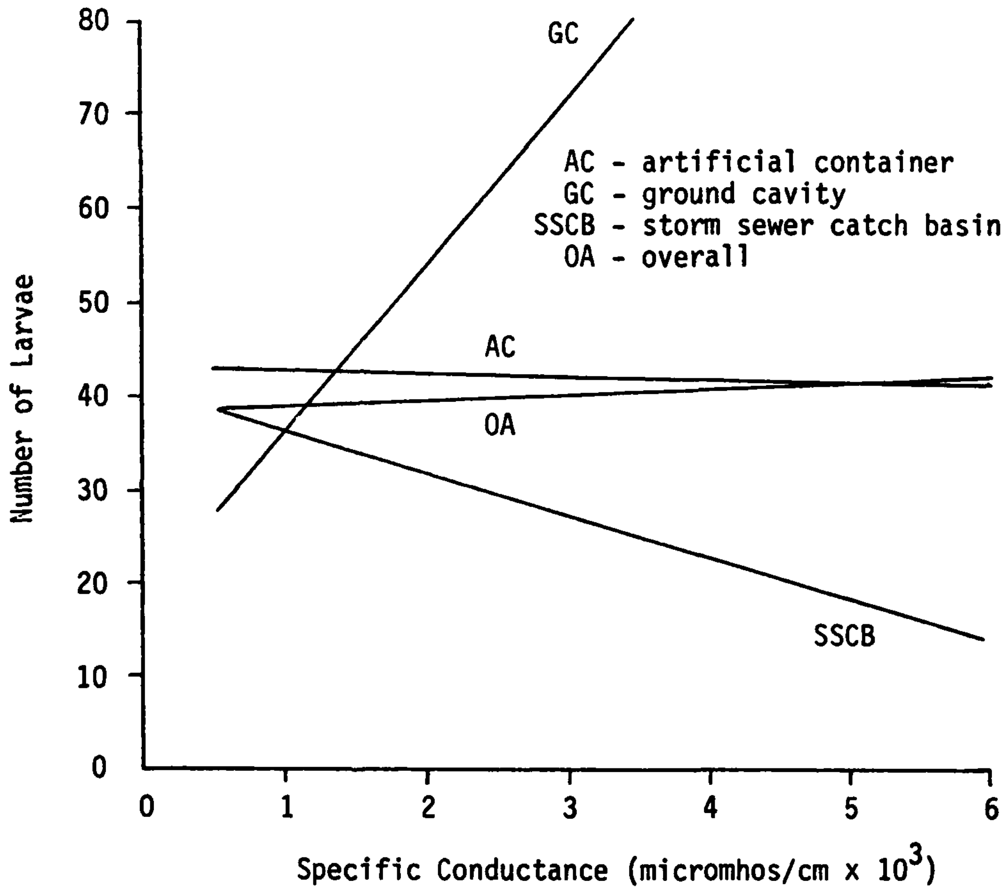


Figure 6A. Regression line comparison of the effects of specific conductance on overall and habitat type larval development.

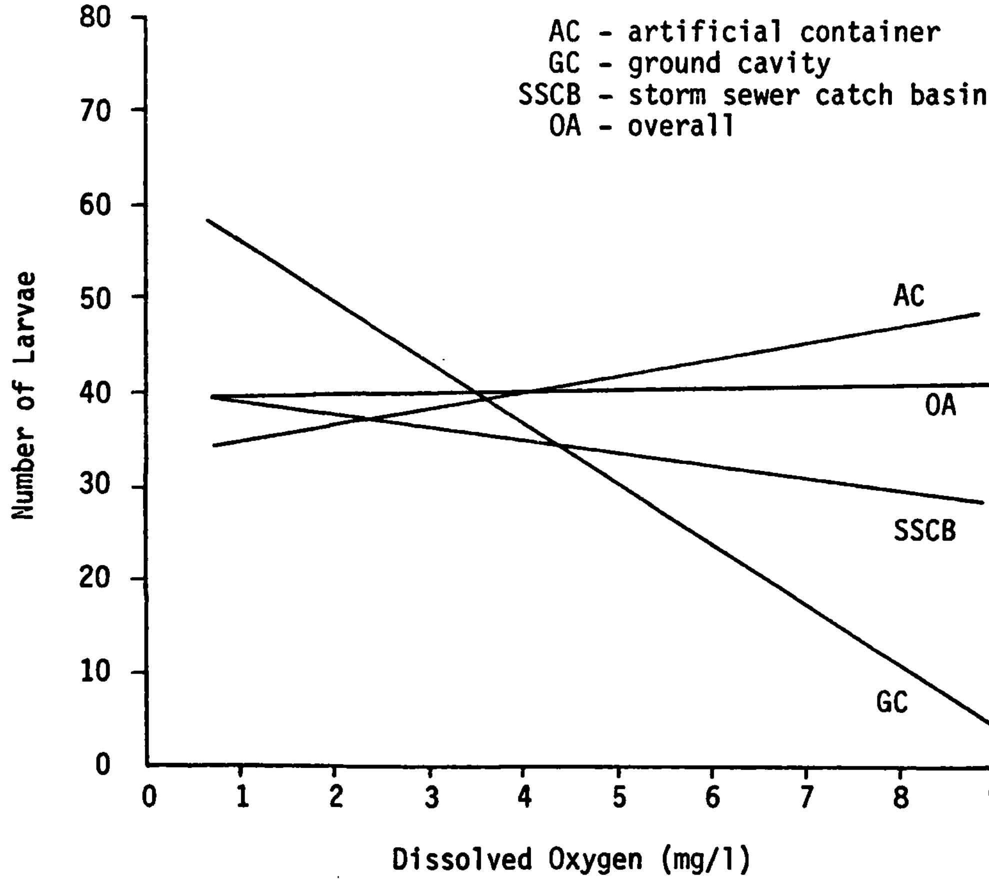


Figure 7A. Regression line comparison of the effects of dissolved oxygen on overall and habitat type larval development.

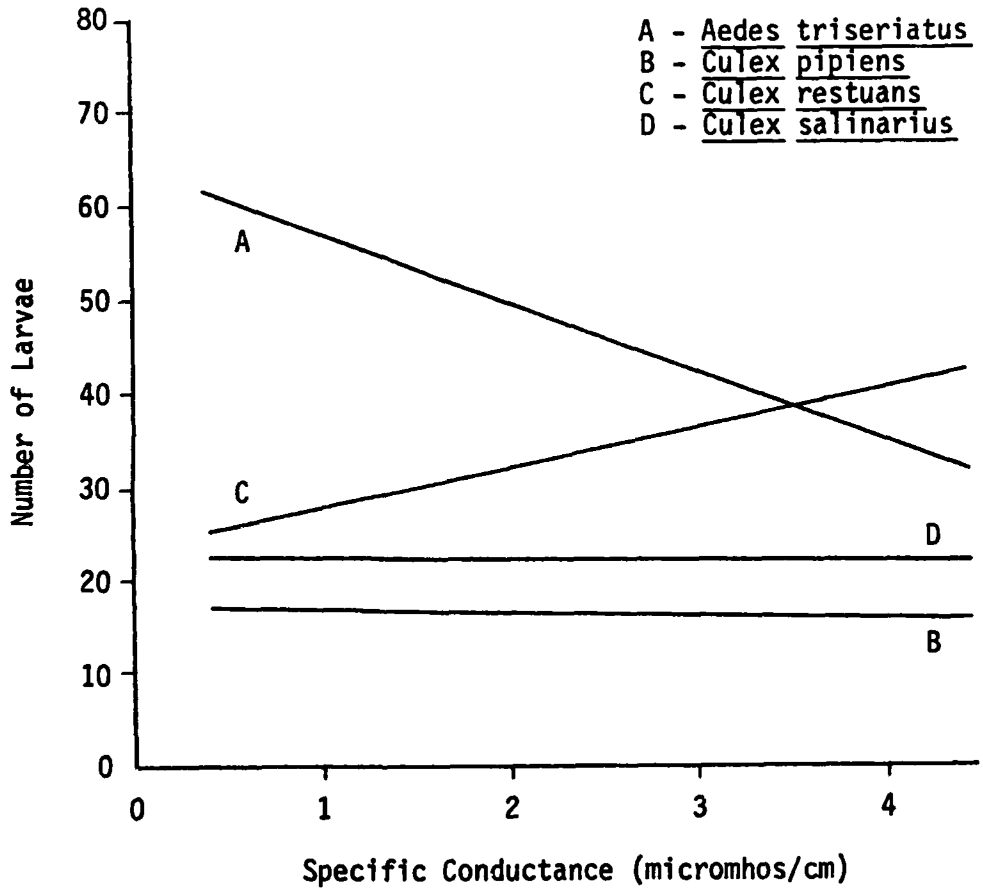


Figure 8A. Regression line comparison of the effects of specific conductance on individual species development.

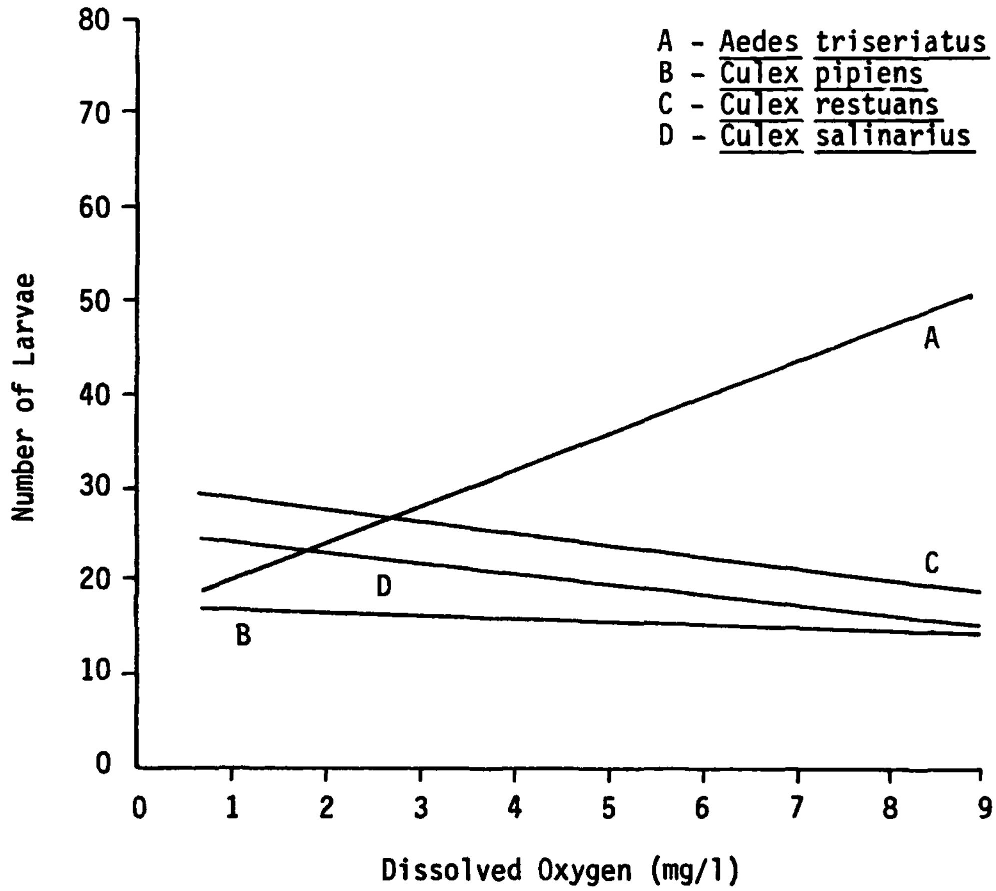


Figure 9A. Regression line comparison of the effects of dissolved oxygen on individual species development.

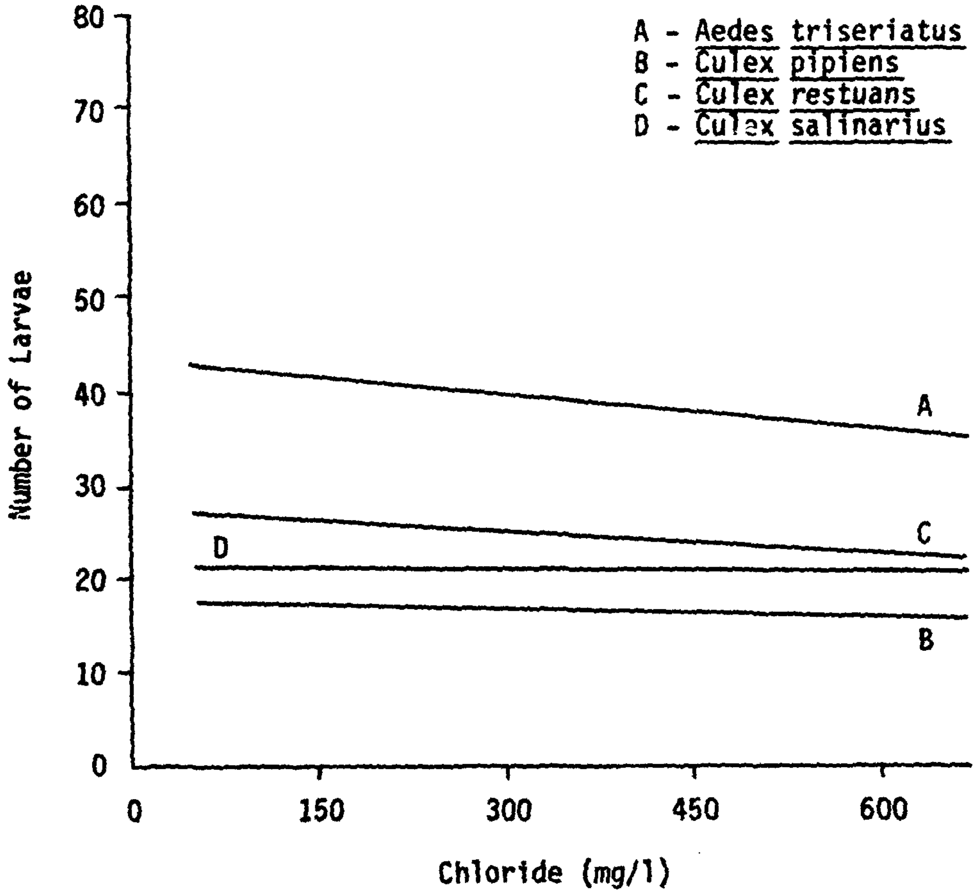


Figure 10A. Regression line comparison of the effects of chloride on individual species development.

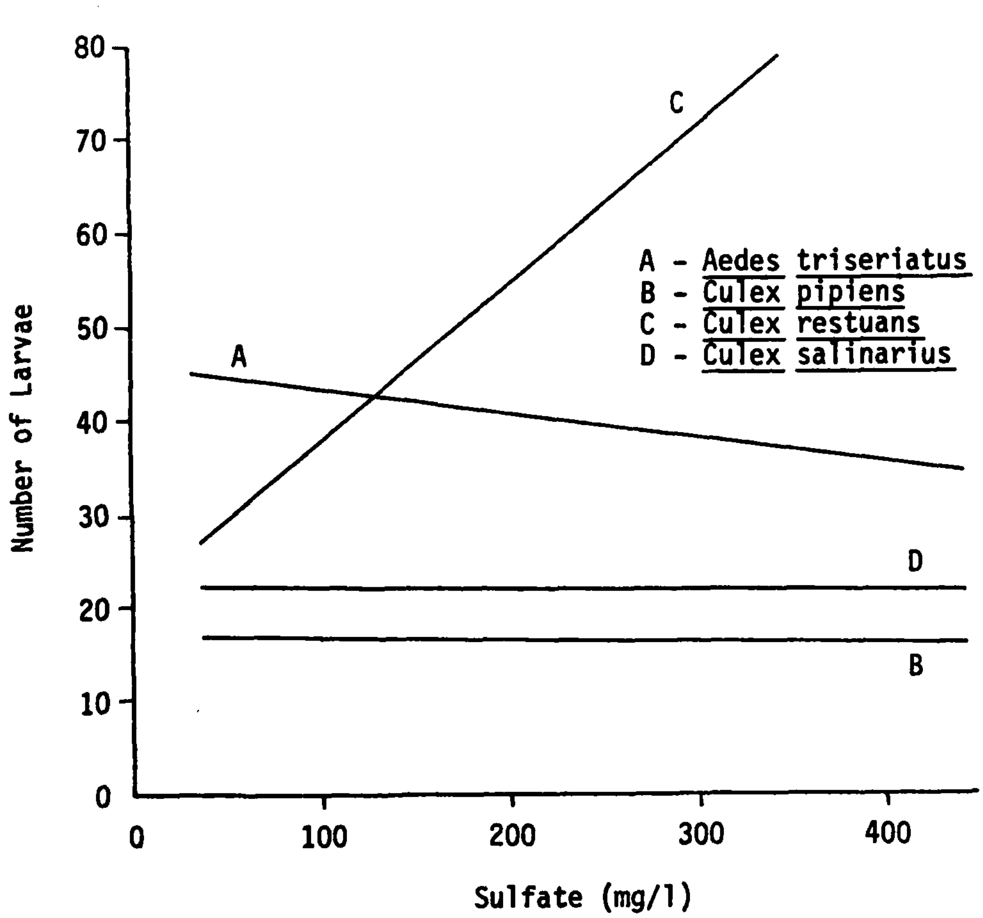


Figure 11A. Regression line comparison of the effects of sulfate on individual species development.

6-8

DT